FINAL CASE REPORT: EgNEFL0603

SPECIES: North Atlantic right whale (Eubalaena glacialis)

DATE: FIRST OBSERVED DEAD: January 22 2006

DATE EXAMINED: January 23 2006

GROSS EXAMINATION LEAD BY:

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HISTOPATHOLOGY EXAMINATION BY:

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RIGHT WHALE NECROPSY REPORT

SPECIES Eubalaena glacialis FIELD NUMBER EgNEFL0603

This report was generated by Michael Moore, Woods Hole Oceanographic Institution, with additional input from Barbara Zoodsma, NOAA; Susan Barco, Virginia Aquarium; Tom Pitchford, Elsa Haubold, Katie Jackson and Alex Costidis from Florida Wildlife Commission; Ruth Frances Floyd, John Roberts and Lisa Farina from U Florida, Gainesville, Mark Dodd and Leigh Youngner, GA DNR and others listed in the attendance log on Appendix page 11. Gross interpretation of the observed lesions was assisted by David Rotstein, U Tennessee. All necropsy images by FWC or WHOI. Assistance with matching the calf to having a mother of #1802 was rendered by Amy Knowlton and Heather Pettis, New England Aquarium, Sue Barco, Katie Jackson and other contributors listed on page 3 to whom the aerial images in the appendix pages 21 to 25 should be credited. Interpretation of the surface defects observed was assisted by Philip Hamilton and Amy Knowlton, New England Aquarium, Jooke Robbins and Scott Landry, Center for Coastal Studies and Greg Skomal, Commonwealth of Massachusetts.

FIRST OBSERVED DEAD LAT 30 14.76 N /LONG 81 03.2 W JAN 22 2006

Fig 1: Ae showing with larg extruding

Fig 2: Impeduncle over dors defect on evident obeached scavenge are prese

Fig 1: Aerial image Jan 22nd showing calf floating left side up with large dorsal laceration with extruding small intestines

Fig 2: Image from boat of dorsal peduncle showing encircling defect over dorsum. The extent of this defect on the right side was not evident once the animal was beached as it had been shark scavenged by that time. Cyamids are present



Fig 3 View of head and dorsal defect with small intestines exuding to animal's left side



Fig 4 View of left peduncle (Jan 22). The diamond pattern is more obvious at this time in comparison to when examined a day later on the beach after further shark scavenging.

IDENTIFICATION OF CALF

Table 1 Analysis of a suite of aerial images show this dead calf to be the same at that associated with mother #1802.

The following sighting record for #1802 was supplied by the New England Aquarium.

<u>Date</u>	<u>Time</u>	<u>Lat</u>	<u>Long</u>	Comment	Sighting
30-Dec- 05	1334	31.58667	-81.02400	With calf headed south. Shark bites on calf peduncle but no linear scars evident	agency WT-SC
8-Jan-06	1254	30.49568	-81.23299	With calf, slow surface swimming and apparent nursing. Linear scars evident.	FWRI
9-Jan-06	950	30.748	-81.28600	With calf	NEA
11-Jan- 06	1659	30.28378	-81.14314	With calf	NEA
21-Jan- 06	952	30.245	-81.07500	Without calf, breaching, blowing bubbles, obvious change in directions, diving and surface swimming	FWRI
22-Jan- 06	1241	30.17103	-81.23804	Without calf, flippering, swimming steadily in one direction, associated with dolphins	FWRI
23-Jan- 06	1229	30.57415	-81.30178	without calf	NEA

WTSC = Wildlife Trust; FWRI = Fish and Wildlife Research Institute; NEA= New England Aquarium,

Images showing the logic behind the above match are shown on Appendix pages 21 through 26.

NECROPSY LOCATION Buck Island Jacksonville FL JAN 23 2006 GENDER FEMALE TOTAL LENGTH 560 cm

HISTORY:

Jan 22 First reported by citizen to USCG. Relayed to Barb Zoodsma at 1030 who called Tom Pitchford and Katie Jackson at FWC.

1105-1201 Photodocumented by FWC Aerial Survey (FWC Images 9002-9070) 1115 Jackson called Zoodsma to confirm a right whale.

1541-1655 Photodocumented and tagged by FWC Boat Survey (Wong FWC Images 8182-8225). Andy Garrett (FWC) reported no line on the animal at time of tagging. Blair mase (NOAA) arranged for a tow by Moran towing. Underway and in tow by 2300. Jan 23. Anchored in St John River at Block Island at 0230. First examined 0715 by Sue Barco and Michael Moore.



Fig 5 Dorsal View



Fig 6 Ventral View

Dried out at mid tide, with FWC anchor line from a tree tied loosely around peduncle. Cutting in to insertion with ragged skin tears, was a ½ inch diameter 25' length of brown colored line with a whipping on one end, and a shredded piece of 3/8 white line both knotted around the peduncle, presumably the residue from tagging and towing given no line seen on animal at time of tagging by FWC team.



Fig 7 View of tag and tow line on cyamid covered peduncle at dawn Jan 23.

Using a line attached to the under flipper, the animal was then rolled on to its back to allow examination of the belly before it was dragged up the beach.



Fig 8 Ventral view after rolling on to back.



Fig 9 Ventral view of throat. Left axillary epidermis is sloughing away



Fig 10 Ventral flukes. A shows two linear incisions, B shows a burst post mortem skin blister. C shows a somewhat eroded fluke notch, D and E show mild linear abrasions.

The animal was then rolled again to place it over three lifting straps to allow removal up the beach without damage to the dorsal defect



Fig 11 Hauling off beach

Note: NE = Not examined NVL= No Visible Lesion

EXTERNAL EXAM:

Two major abnormalities were encountered, a dorsal defect, and a complex laceration pattern on the peduncle. To track samples of these regions, sampling stations were each assigned a letter, which were recorded on sketches of the region in question. Each sample was labeled with its source station letter. Those sketches can be found on Appendix pages 4 though 7. Refer to them, along with Appendix page 10, where the samples taken are all listed. The data sheets were recorded by Leigh Youngner, GA DNR, and the sample sheet by Mark Dodd GA DNR. These sheets are included as scans to avoid transcription errors and to expedite report production. Original data sheets are archived with the author at

WHOI, should the need arise for clarification of specific data entries. Photographs of the sample sites, and samples taken are all logged on the photolog sheets. Thus to fully understand the provenance of a particular sample submitted for histology, refer to the photolog for images showing the slices and the source site, and to the sketches for the station location. The umbilicus was a ring of epidermis overlying granulation tissue. See image right.



Umbilicus

DORSAL DEFECT



The dorsal defect was an irregular opening centered over the dorsal midline, with intestinal remnants draped over the left axilla. The defect was 2.4m x 1.2m (measurements were estimated from photographs).

Fig 12 View of Dorsal



Defect

Fig 13 View of rostrum and cranial part of dorsal defect



Fig 14 Dorsal Defect Station A. This portion may be a shark scavenge bite.



Fig 15 Dorsal Defect Station B



Fig 16Dorsal Defect Station C – note the apparent clean cut at this point



Fig 17 Dorsal Defect Station D – note this appears to be a ragged 'burst' rather than a cut.

The edges of the defect were mostly smooth cut, with a gray shaggy appearance. The outline was mostly ragged at the cranial and caudal portions, and more straight cut in the middle on both sides.



Fig 18 Dorsal Defect Station E



Fig 19 Dorsal Defect, including entire right edge (top of image). Illustrating stations O and N. This image was taken after the animal was dragged up the beach and then lain on its left flank, as whilst on the beach the right edge was obscured by overlying tissue. The images of this defect shown above this image were all taken while the animal was lying

on its right side on the beach. It was not possible to ascertain if the two sides of this defect could accurately re-appose, given the extreme deformation of the carcass by the pressure of the protruding tissue, however the shapes of the tears at the cranial end at least suggest that this was somewhat possible.

PEDUNCLE

The peduncle was covered in cyamids, as illustrated on page 5 above. These were removed to allow examination of the underlying skin surface. The insertion of the flukes had a ragged encircling laceration within which the ropes used for tagging and towing were embedded as shown on page 2 above. There was no tissue reaction below this laceration. Anterior to this along the lateral aspect of the left side of the peduncle there was a complex series of defects through the skin. The surface pattern ranged from straight lines wrapping around the left half of the peduncle and over the dorsum down towards the shark bites on the right and along the length of the animal, to vee shapes, to diamonds. These ranged in depth from mere impressions to full depth of the epithelium of ~12mm. They did not penetrate the dermis. The deepest defects stopped at the epidermal/dermal junction. Some showed raised epidermis, others simple vee notches. Sections though these areas revealed a consistent underlying dermal lesion in the form of an inward pointing triangle, with the base at the epidermal/dermal junction. The edge of this lesion was sharply demarcated. The color was red, suggesting a peri mortem/ ante mortem host response. The timing of this lesion awaits histological investigation. The color of the cut surface of the epithelium varied, but in some regions was white especially near the base, and enlarged, suggesting a physiological response to the inciting external source.



Fig 20 Left peduncle. Towing laceration at extreme right of image (see oval). Remainder of defects appear to be pre mortem caused by a fine cutting edge such as monofilament. Shark scavenging is evident on the dorsal mid line in the above image (arrow). The image of the peduncle shown at sea on page 2 should be compared here to show what was removed by sharks.

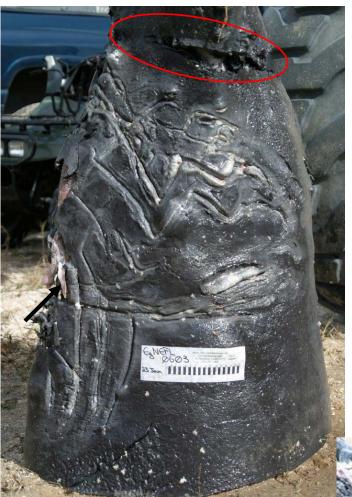


Fig 21 Left and below – left peduncle. Diamond patterns measured 13x9 cm, 12x10 cm, and 12x10 cm. between opposing apices. Red oval shows towing laceration. Shark postmortem scavenging shown, with black arrow at left.

Fig 22 Close up of left peduncle



These linear defects were sampled histologically at a number of stations.

Station P



Fig 23 General location of station P: right of dorsal midline on a defect partially encircling the peduncle.



Fig 24 Station P Sample site

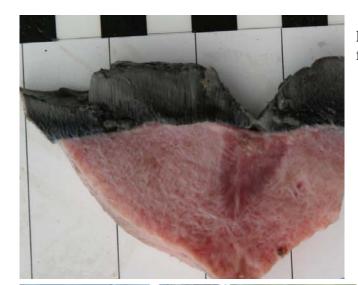


Fig 25 Station P – Sample as preserved for histology



Fig 27



Fig 28

Fig 26a Left Peduncle Cuts for sample stations H1 and H2. Arrow points the defect sampled in H1 (Fig 27) and, knife to that samples in H2 (Fig 28).



Fig 26b Sample taken from station H1

Right peduncle

Fig 29 There were two crescentic lacerations evident on the right side. Their shape was characteristic of shark bites. They measured 22cm across the gape. No shark teeth could be found in the wounds.



The caudal most bite was the deepest. The dorsal mid line, shown at the base of the above image, and the area between the two large bites had a series of smaller bite wounds and nibbles. On the basis of the boat images above showing an intact peduncle ridge on Jan 22, these latter wounds were presumed to represent shark scavenging in contrast to the two larger premortem predation bites shown here, both of which are evident on the aerial image taken on Dec 30 see Appendix page 25. Histology was sampled from Stations H1, H2, H3, P, Q, R see Appendix page 6 and photolog.

INTERNAL EXAM:



Figure 30 Transected peduncle. To obtain a better examination of the peduncle it was removed from the body. A normal inter-vertebral space was observed. The dorsal third of the musculature was pale in comparison to the rest of the section, suggesting that the dorsal orientation when floating had resulted in dependent lividity.



Fig 31 The right blubber coat was removed to reveal similar ventral lividity with dorsal drainage, with reddened edematous material at the subdermal sheath. None of this discoloration extended in to the musculature or the blubber. Some areas were mottled green, and the tissue contained numerous small gas bubbles. It was sampled for histology, but assumed to represent post mortem dependent lividity.

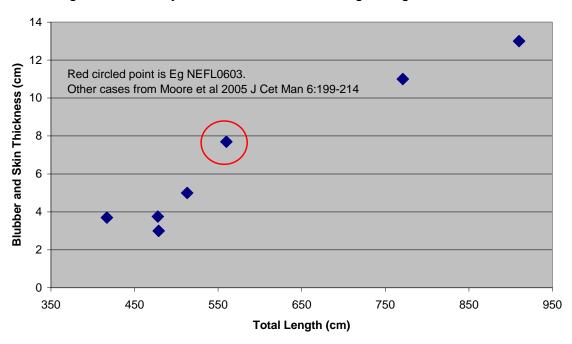


Fig 32 Dorsal Axillary Blubber Thickness vs Total Length in Right Whale Calves

DIGESTIVE SYSTEM

Mouth: NVL

Baleen: Left baleen 222 plates. Right baleen 231 plates. All sampled and frozen at GA

DNR.

Tongue: Vee notch present in dorsal surface. Three histology stations sampled. See image at right.

Esophagus: NVL 2 sites sampled for histology

Fore Stomach, Main Stomach, Pyloric Chambers:

Empty – moderately distended with gas. Mucosa autolysing

Duodenum: NE

Intestine: NE

Colon: Empty NVL

Anus: NVL

Describe food-stuffs found in GI Tract: None

Parasites found in GI Tract: None

Liver: Normal - autolysed

Pancreas: NE

Mesenteric Lymph node: Possibly sampled.

BLUBBER:

Thickness normal, no discoloration except as described for peduncle defect lines in superficial dermis above.

MUSCULOSKELETON

NVL



Fig 33 Tongue as sampled

Axial Muscle:

Red tinged fluid in the mid lumbar area

Axial Skeleton

Thoracic, Lumbar, Caudal Vertebrae: NVL – epiphyses, and neural arches to each vertebra not fused.

Mandibles NVL



Figure 34 Mandibles

Flippers: The left scapulo-humeral joint was grossly normal.

Ribs & Sternum: NVL

Fig 35 Ribs



Flukes: Small (~2-3cm diameter) round erupting lesion on the dorsal aspect of the left fluke, approximately half way between the fluke insertion and distal most tip associated with post mortem blistering. Two similar lesions ventrally, along with mild impressions from bilaterally from fluke insertion to notch (see Fig 10)

Cranium: Brain autolyzed. Both ears removed and frozen.

THORAX

There was a large amount of blood-tinged liquid within the sub-dermal sheath in the ventral and ventrolateral aspects of the cranial thorax, consistent with lividity. A sample of the sheath with associated hypaxial muscle was collected for histopath. A sample of the diaphragm was collected for histopath.

CIRCULATORY SYSTEM

Heart:

Pericardium: c 500ml re fluid – probably result of autolysis. Measurements of the right ventricle: apex: 5 cm (diameter of chamber?); adjacent to the right AV valve 7 cm (diameter of chamber?); the right AV valve was 8 cm in diameter.

Great Vessels:

Pulmonary artery: There was a large clot in the pulmonary artery. It was measured and photographed (> 12 inches long). The wall of the pulmonary artery was 1 cm thick (measurement taken 9 cm proximal to the pulmonary valve). The internal diameter of the vessel at this location was 6 cm.

Aorta: The thickness of the wall of the aorta was 1.2 cm (measurement taken approximately 15 cm from valve). At this location the diameter of the vessel was 6 cm.

The ductus arteriosus was open (not closed and probe patent) but constricted and on its way to closure as suggested by the much larger diameter of the pulmonary arteries, and the diameter was 1.5 cm. Photographs taken.

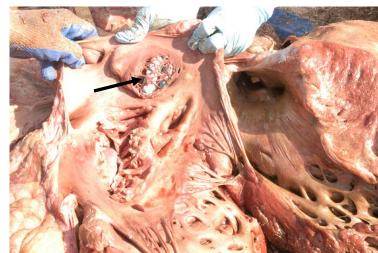


Fig 11 Foramen ovale -

The foramen ovale was partially patent and spanned by a fenestrated membrane. The diameter was 7cm, See Fig 11 with arrow.

Cranial Circulation: NE

Spinal Circulation: No epidural clots were seen at the occipital and caudal vertebral sections examined.

RESPIRATORY SYSTEM

Blowholes: NVL

Nares: NVL

Larynx: NVL

Trachea: NVL

Lungs: No adhesions or fibrin present in right pleural space. Both lungs appeared properly inflated with no apparent atelectic regions. No cysts or nodules were observed within the parenchyma. Both lungs were pale tan on serosal surfaces, dark red and slightly bloody on cut surfaces.

Bronchi: NVL

Odd structure found in the mediastinal area: Cylindrical, red-colored structure was found which is believed to have been cartilage. The structure was located adjacent to the airway, in the stroma between the trachea and the aorta. The structure was examined by Drs. Moore and Roberts and Mr. Costidis. A sample was taken to UF (in formalin) but it was not divided into three samples so that it could be kept intact as an aid in identification if that proved necessary.

URINARY SYSTEM

The umbilical arteries were open from the bladder tip caudad, but closure could not be determined cranially due to the extent of decomposition. There were well-formed, loose blood clots within both umbilical arteries in the region of the bladder. The urachus appeared closed at the bladder tip but the extent of decomposition precluded examination craniad.

Kidneys: Small remnants possibly found. Sampled for histology

Ureters: NVL

Bladder: NVL

Urethra: NVL

ENDOCRINE & HEMOLYMPHATIC SYSTEMS

Thymus: NE

Thyroid: Right grossly normal

Parathyroid: Sampled (identity in question)

Adrenals: Possibly sampled. Autolysed

Pituitary: NE

Spleen: NE

Lymph Nodes: NE

REPRODUCTIVE SYSTEM

FEMALE. Nulliparous female, as supported by total body length, as well as transparency and lack of vascularization of broad ligaments of the uterus.



Opened vagina, cervices and uterus with both ovaries

Ovaries: NVL autolysed

Uterine Horns: NVL

Uterus: NVL

Cervix: NVL

Vagina: NVL

Mammaries: NE

CENRTRAL NERVOUS SYSTEM

Brain: Autolyzed

Spinal Cord: NE

SENSORY ORGANS

Eyes: NVL Right eye frozen. Left eye fixed in 10% NBF.

Ears: NVL Both frozen

PRELIMINARY GROSS DIAGNOSES AND INTERPRETATION

Diagnoses are preliminary and based upon gross interpretation of findings. Histopathologic examination of tissues is pending and may lead to diagnosis amendments or addendums.

Preliminary Diagnoses:

Integumentary System:

A. Dorsum: Regionally extensive epidermal defect with cranial and caudal irregular and roughened edges and segmental smooth edges without associated subdermal edema or hemorrhage.

B. Peduncle:

- 1. Regionally extensive epidermal and dermal indentation and penetration with overall pattern formation of diamond, vee, and straight lines (net entanglement, vs. shark bite, vs. both) with localized tissue reaction.
- 2. Multiple semi-circular cutaneous defects (shark bite)
- 3. Cutaneous Cyamidiasis, severe.

PRELIMINARY INTERPRETATION

The cause of death is open at this time pending histology.

Peduncle

Tissue reaction observed in peduncle at site of integument indentation, deformation, and loss indicating ante mortem host response. The various patterns of vees, straight lines, and diamonds as well as distance of separation of individual shapes is most suggestive

(consistent) with man-made materials (e.g. net). It could be that all the defects on the peduncle were induced solely by shark predation and scavenging, however the diamond pattern in particular is hard to conceive in this light. Review of these lesions by whale and field shark biologists has given to mixed opinions in this regard, however the weight of evidence would suggest that the linear scars near the peduncle were entanglement induced, whilst those further forward, with less of a regular pattern were shark induced. Images pertinent to this discussion, both from aerial survey of this animal whilst alive, and other relevant precedents from previous cases are included in the appendix (pp 27-33). An opinion from the Massachusetts State Shark Biologist is rendered in favor of entanglement being involved in addition to shark damage is shown on Appendix page 34. Ante-mortem migration of cyamids to the peduncle may further indicate an ante mortem host response. Their concentration at the peduncle may be the result of the animal being slowed down by the lesion (debilitation) or microhabitat generation by the presence of an external structure (e.g. netting). Wedge-shaped discoloration within the dermis subjacent to regions of epithelial indentation likely represents sites of host response. This could include vascular compromise, thrombosis and subsequent infarction; inflammation and repair; or inflammation associated with an infectious process.

Dorsal Defect

The dorsal defect was characterized by regions of ragged/irregular epithelium and smooth edges. Hemorrhage and edema were unassociated suggesting that this was a post-mortem occurrence. The cause of this defect is not known. The following is not an exhaustive list of causes, but are prevailing differentials at the time of this report's preparation. Additional analyses may yield new differentials. Basically, external or internal forces could explain the observed changes. Internal would include natural decomposition and gas production leading to excessive internal forces and subsequent venting. Ragged edges at the cranial and caudal region could support such an explosive event, but do not explain the smooth and staggered edges observed. External causes of venting include shark scavenging or cutting object. Irrespective of the scenario leading to this unusual lesion, this was not grossly evident from available aerial survey images that would suggest that this lesion occurred post-mortem. Opposability of the lesion could not be determined.

Other pertinent observations

It is worth considering the available information to assess potential scenarios that led to this mortality, on the basis of what we now know.

- 1. The blubber thickness would suggest that the animal was in good body condition when compared to other right whale calf mortalities see Fig 32.
- 2. The available gastrointestinal tract was empty. Gastric milk and colonic contents have been observed in other dead right whale calves (Moore et al 2005), thus it seems likely this animal had not suckled for a day or more prior to death.
- 3. The linear defects observed around and on the left side of the peduncle appeared to penetrate as far as but not beyond the base of the epidermis. Had these lesions been shark induced there would have been a more variable depth of penetration beyond the base of the epidermis, as seen on the right peduncle. This was not the case as illustrated by Fig 26b. Shark bites would also have been more punctuate.

- Thus the assumption that these lesions were induced by wraps of monofilament net, perhaps streaming under the ventral flukes appears more likely.
- 4. The spacing between the linear defects on the peduncle were a) at times consistently spaced, and b) more widely spaced than any known adjacent shark teeth.

Thus the weight of evidence is that the peduncle had pre mortem lesions associated with pre mortem shark predation and monofilament net entanglement and post mortem shark scavenging.

APPARENT SCHEDULE OF EVENTS

This animal was observed with its mother #1802 off St Catharine's Island GA (68 nm north of Jacksonville) on December 30^{th} 2005. At that time it only had shark bites on its peduncle. It was observed with linear scars, possibly as a result of entanglement (see discussion in Appendix pp 26-34) on the peduncle in addition to the shark bites on January 8^{th} 2006 12 miles north of Jacksonville. No entangling material was ever seen. The calf was last seen alive on January 11^{th} . Thus it appears the linear scars (?entanglement) occurred at least 14 days prior to discovery of the carcass on Jan 22^{nd} . The calf died somewhere between Jan 11 and Jan 21^{st} . Thus it was dead between 2 and 11 days when discovered on the 22^{nd} , and it was probably entangled for between 3 to 21 days before it died.

Station 41008 - GRAYS REEF - 40 NM Southeast of Savannah, GA http://www.ndbc.noaa.gov/station_page.php?station=41008

Michael Thore Date: February 1st 2006

shows a surface water temperature for recent days of about $60 \,^{\circ}$ F ($16 \,^{\circ}$ C). This water temperature could lead to a body condition as observed in this carcass in about 3 to 6 days.

USF Institute for Marine Remote Sensing

http://imars.usf.edu/cgi-bin/db?site=east&index=1&type=st&mode=runmean
List the SST for Jan. 22, 2006 composite satellite image at 15.75 ° C (60 ° F) on 1/19/06
(the nearest day with the least cloud cover) at Latitude Longitude 15.83 30.2329 81.0411

Signed by:

Michael J Moore, Woods Hole Oceanographic Institution, Woods Hole MA 02543

NECROPSY CREW

In no particular order **VAQS** Sue Barco

FWC

Arthur Wong **GA DNR**

Jimmy Hughey Leigh Youngner Alex Costidis (MMPL) Mark Dodd

Andy Garrett

Katie Jackson UF

Tom Pitchford Carla Phillips Leslie Ward Carolina Ruiz Elsa Hubbard Ruth Floyd Lisa Farina John Roberts

NOAA

Barbara Zoodsma

Don Lewis **WHOI** Michael Moore

Acknowledgement

Although this report was authored by Michael Moore, this document represents the labors of a vast number of people who contributed images, time, materials and energy in to this event both on the beach, and in analyzing the resultant material.

Supported NOAA Contract # NA04NMF4720392 to M Moore and W Mclellan and a subcontract to Virginia Aquarium, in addition to resources from FWC and GA DNR.

Conducted under NOAA Permit 932-1489 to Dr. Teri Rowles.

PHOTOGRAPH LOG

See handwritten logs on Appendix pages 11-20.

In summary: File number range. Subject. (Photographer)

FWC:

9002-9070. Aerial Images Jan 22 8182-8225 On water images Jan 22. (Arthur Wong) 8240-8499 Images at Necropsy. (Katie Jackson)

MOORE WHOI SONY:

7282-7354. Images at Necropsy. (Sue Barco and Michael Moore)

Evaluation of Human Interaction
Field Number EgNEFL Species Eubalaena glicialia Date 2/22/06 Code 1 2 3 4 5
External Examination A. Body Condition: Emaciated specimens often exhibit sunken epaxial musculature and neck Emaciated Not Emaciated CBD N/E
B. Net or Line Marks: Indicate Y/N/CBD/NE for each area and carefully describe net or line marks: Head D.Fin L.Flipper R.Flipper Peduncle Other
C.Fishing Gear Present on Animal (Yes) or (No)
D. Gear Retained (Yes) or (No)
E. Penetrating Wounds: Yes No CBD N/E
F. Mutilations : Body Slit or Mutilated? Yes No CBD N/E
G. Hemorrhaging / Bruising: Yes No CBD N/E Describe extent and area: hemorrhaging was found in association with peduncle lacerations photos and Hissues collected for histology section H, P, R, Q Internal Examination A. Sub-Dermal Hemorrhaging: Yes No CBD N/E
Describe extent and area:
B. Broken Bones : YesNoCBDN/E Describe :
D. Stomach Contents Retained : Yes V No
E. Histopathology Samples Retained : YesNo
F. Gross Pathology : Yes / No CBD N/E Describe :

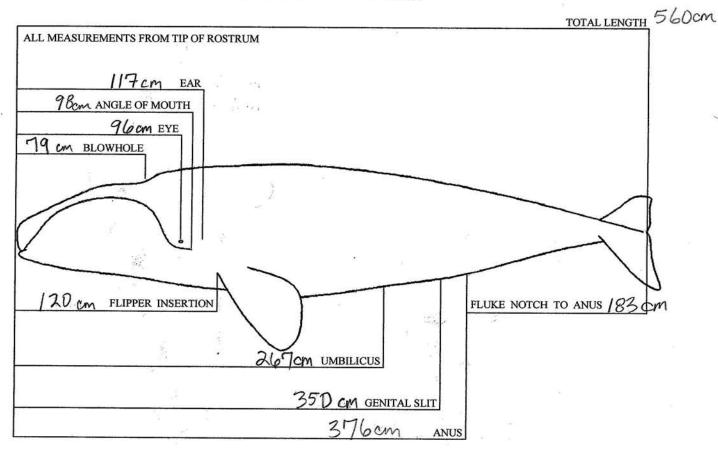
^{*}CBD - Can not be determined

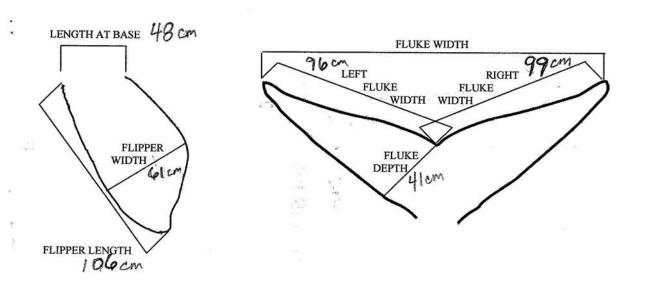
^{*}N/E - Not Examined

EGNEFL O 603

Right Whale External Morphometrics

WRITE MORPHOMETRICS ON LINES

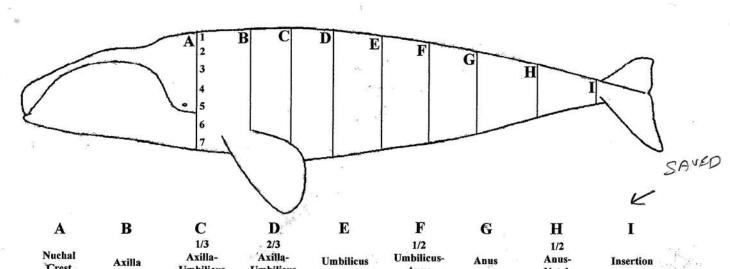




leigh Youngher @dur. state.ga us one Construction Way Bruswick, 64 31520

RIGHT WHALE BLUBBER THICKNESS

Field # EgNEFL 0603 Side Examined Right Observer _____ Date 2/23/06



109/7.0 2.0/5.7 NT NT NT NT 1.5/9.5 N/T

2 N/A/8.0 1.5/5.1 1.4/4.4 1.7/5.2 1.6/5.7 1.4/29 2.0/8.3 N/T

1.2/6.5 NAS.0 1.5/5.4 1.9/2.01.9/6.0 1.9/6.2 2.5/5.3 2.0/2.0

10/6,5 1,5/6,9 2,0/6,8 2,0/6,5 2,4/2,0 1,9/8,2 2,3/5,0 2,0/8,0

2,0/9,0 NP/7.5 2.1/7.0 2,0/7.52.1/7.0 20/8,1 2.0/5.0 2.0/8.1

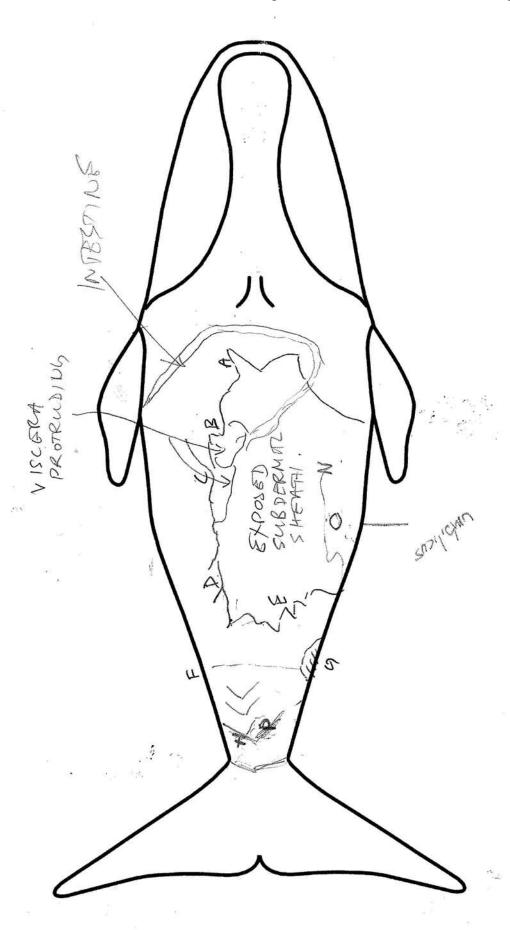
1.5/7.3 4/8/8.0 1.8/8.5 2.0/6.92.0/7.5 21/8.1 23/5.0 2.0/8.0

NT NT NT 1.5/3.5 1.5/2.02.0/8.2 2.0/8.0 2.0/6.1

Ventral

Circumference with blubber NOT TOKEN - FOO BLOWN OUT

NOT TAKEN Circumference w/out blubber



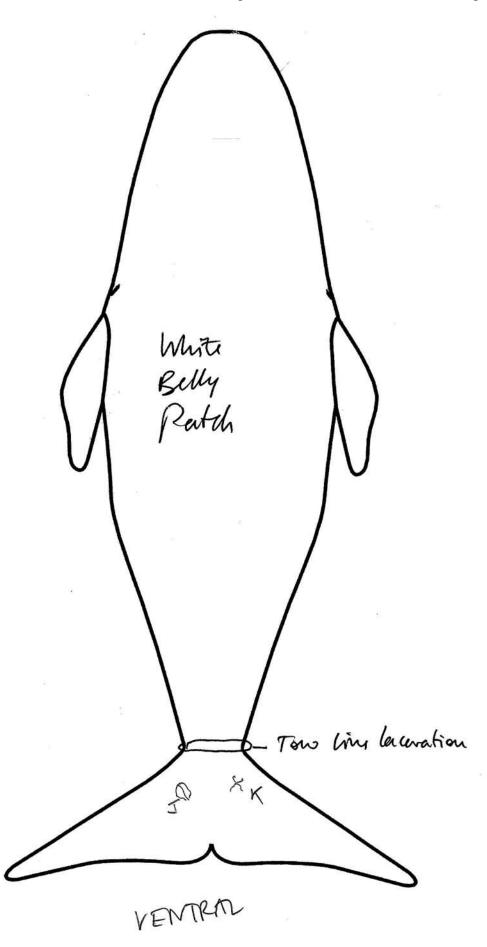
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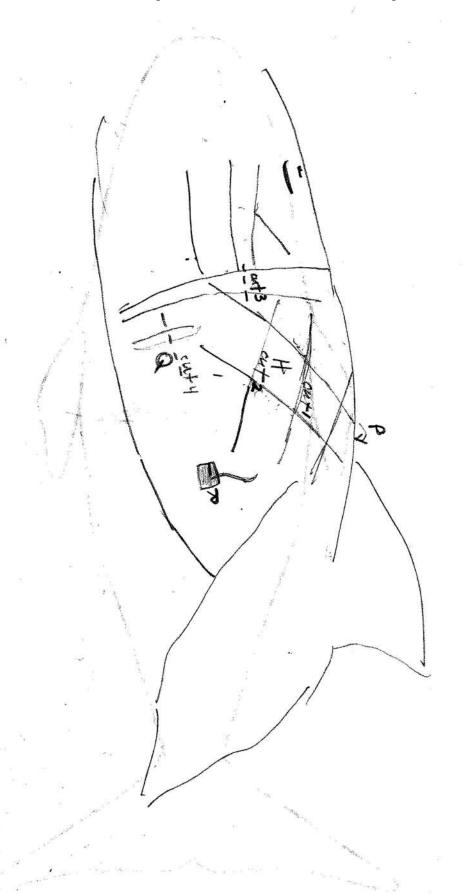
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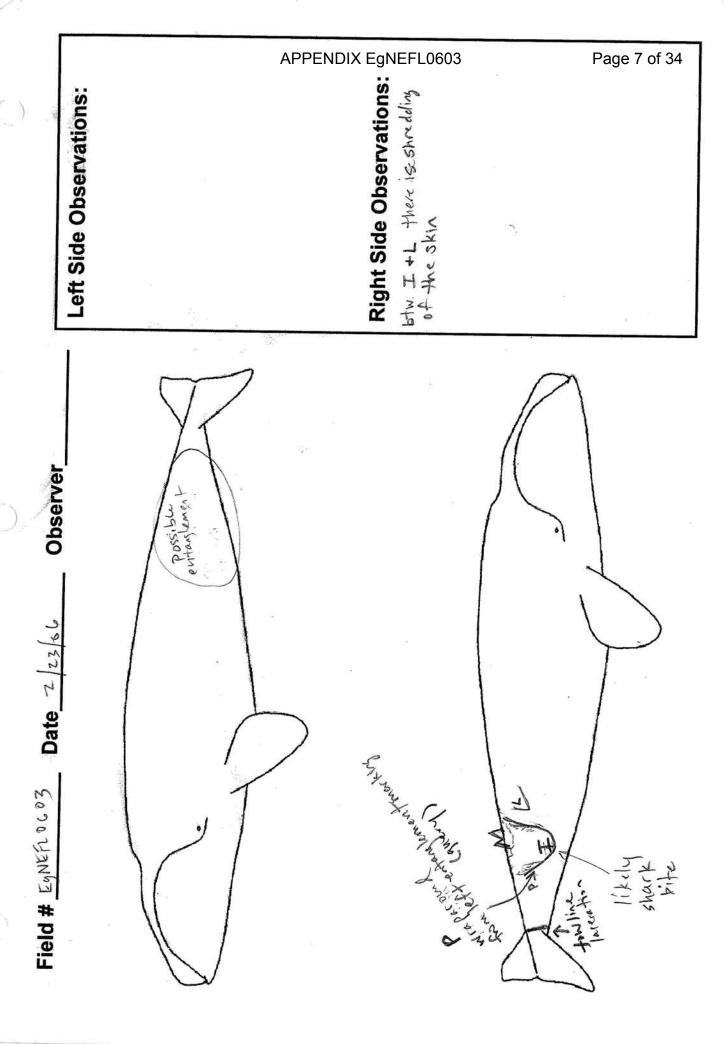
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GONEFL 0603



left peduncle





APPENDIX EgNEFL0603 Page 8 of 34 Right Side Observations: Left Side Observations: Observer アント Field # NEFL 66.03 Date 1-23- 66

APPENDIX EgNEFL0603 Page 9 of 34 Right Side Observations: Left Side Observations: Field # EgNEPL Chos Date 1-23-100% Observer

Histopathology Checklist

-	Blubber/Epidermis	Lung		1
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	Abrasion sites	Left Atrium		
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V	Station B(Doisil Cac)	Right Atrium	10	
V	Station C (Dorsal Lac)	Right Ventricle	1	
11	Station D (Dorsal lac)	Aorta	1	
V	Station I	Pulmonary Artery	1	
V	Station E	Esophagus	1/	1.00
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V	Station N	Main Stomach	10	
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People Attending 2/23/06 Name contact # / email Affiliation ArtieWong **FWC** 904 237 197 / arthur. Wong Omythe. JIMMY HUGHEY FWC 904 838 0213, - Jimmy hugher pellout ENC-MMPL 727-423-4494/Alex Costidis & Justine Dady & Costidis & My fuc NOAA SHIP LIAISON 904233-8627 DLEWIS & JANY MY COSTIDIO ORG Alex Castilles DON LEWIS Mark Dodd 912-264-7218/mark_ Dodd e 029 904-573-4918 dar state ga. 45 Georgia DWR Andy Garret Fuc 4 and y gallets Q my Fue. com Katie Jackson FLUC 904-237-4320 Kathe Jackson@muffix Sue Barco VAQS 75615-0740 ocrabe ords com TOM PIRUXORD 777 425 8430 Fwc TOM, PITCULARDE my Fine, com GAMR Leigh Youngner Carla Phillips UF 352-246-6935 danhearl@ufl.edu Carolina Ruiz 352-281-2569 UF caroruizenfledu Rule Ployd 352-538-0606 WF Francis - Florid R appayl vetored Uffeder USA Fama W 352-396-5037(W) farinal @ monil 352-392-4700 x+5814 vermed uffet John Roberts UH 357-3512 4700 X roberts @ mail vet med of loda Looke Ward Fuc 727 8968626 lostie worlding Fire 5/59 Hausold FUC 727 8968626 who Michael Moore 508 289 3278 MM oor Ochoiledu

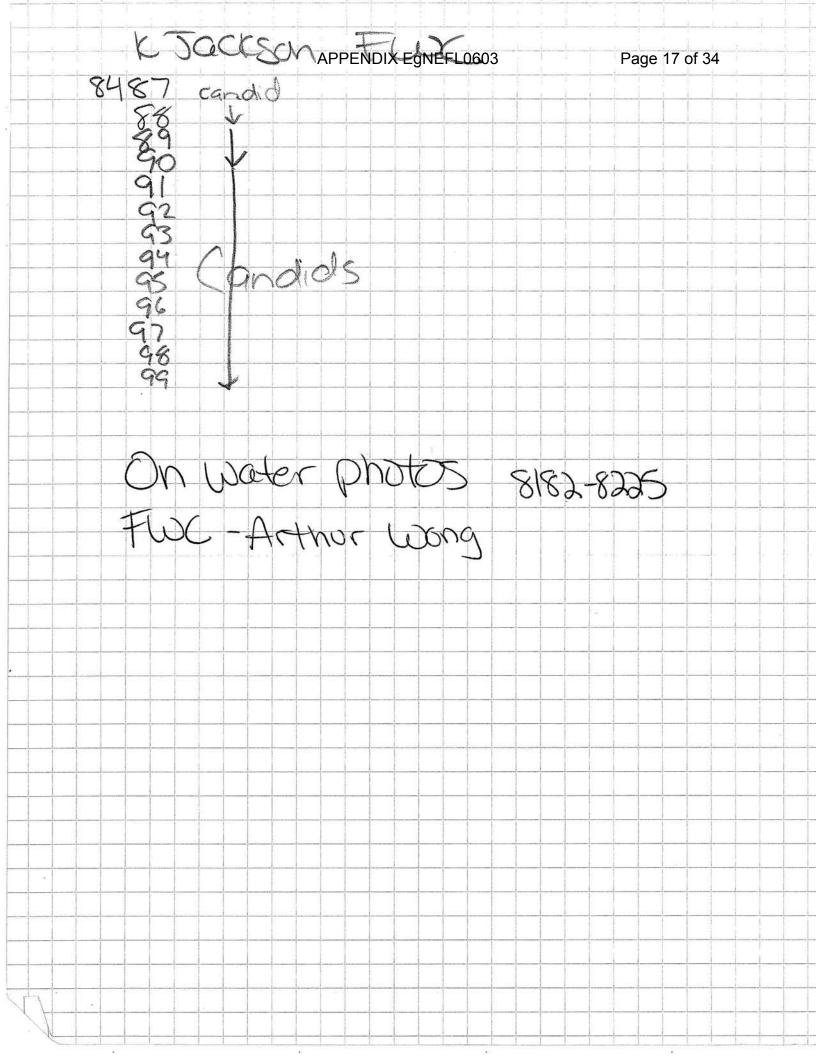
FUC camera Republix EgNEFL0603 Page 12 of 34 full body Rside blubber samples from obrigal lesion
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-WC APPENDIX EgNEFL0603 Page 15 of 34 8407 Sife 08 sample abdomen + where horns + bladder + chine val + foramenoval? /caudal venarava Daleen V thymus? Bladder (Pmpty) Fight lung 7 esophagus 20 1 gorta docsa left thoracia cavity down side 23 24 diaphram distal e sophesus kidney kidnes heart right ventricle 20 30 32 blood clot thon pulmonary arter Liver (Sample) bile duct (Sample) 36 pulmonary artery 39 ascending acrts (Sample) Oscending aorta lect ventricle Uterus 43 ovaries close up ovary

K Jackson Fluc Page 16 of 34 8446 Close APPENDIX EGNEFLO603 47 parasité (Fascia next to trachea) O Ribs Right Side 2 mandibles Uterus/overies (overall) Stroma between tracked t bronchus unidentified Structure 62 sample of unidentified structure 63 trached sample Sy left ventricle right ventricle right papillary muscle alorta (sample) 6% forginen ovalle 70 right attim (Sample) lungs lealing (Sangole) (Cardal left ling) righting (sample) (not over if righting) mediastinal tymph nock (sample) 84 close up ductus arteosis 81 (Sample 82) (Sample 82) Candid 83 Candid 84 Candid 85 Teff (105) (39 mple) left thymus (shadow)



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7264	MIDDLE 1/3
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Images 1 and 2. 1802 and nursing calf. White belly is visible on calf (photo FWRI 08 January 2006)



Image 3. Necropsy image of calf with white belly (23 January 2006)

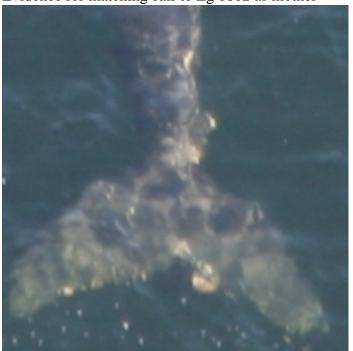




Images 4 and 5. Peduncle and flukes of 1802's calf (FWRI 08 January 2006)



Image 6. Right peduncle of necropsied calf (23 January 2006)





Images 7 and 8. Dorsal peduncle and flukes of 1802's calf. (FWRI 08 January 2006).

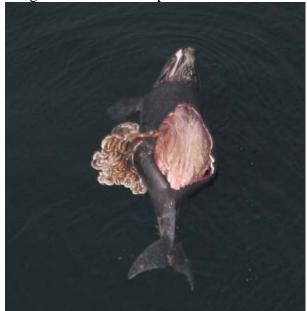


Image 9. Dead floating calf 22 January – note peduncle (FWRI 22 January 2006)



Image 10. Dorsal and left peduncle of 1802's calf (08 January 2006)



Images 12 and 13. Dead calf – note peduncle (FWRI 08 January 2006)

APPENDIX EgNEFL0603



Image 11. Left peduncle necropsied calf (23 January 2006)

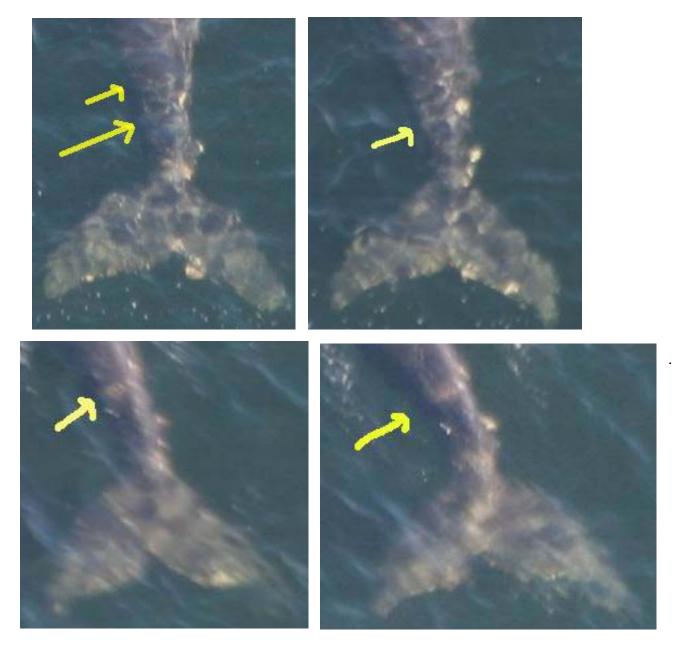




Image 14. Calf of 1802 at first sighting (WT-SC 30 December 2005).

Evidence for matching calf to Eg 1802 as mother APPENDIX EgNEFL0603

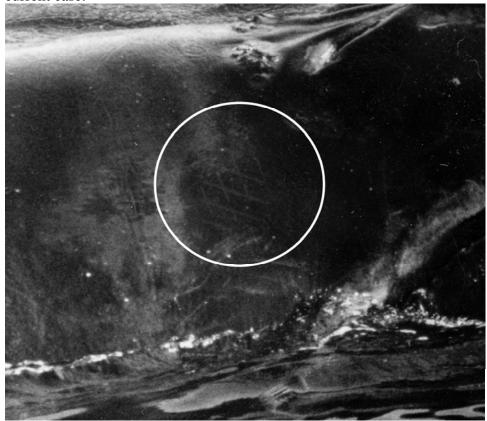
Images taken on January 08 2006 that demonstrate the presence of the entanglement scars on the left peduncle.



BACKGROUND INFORMATION

Pertinent prior images to compare with this case to assist in the interpretation of the observed lesions.

Image of Right Whale #2212 taken by the Provincetown Center for Coastal Studies on 8/10/92. No gear was apparent on the animal at this time. Thus there is precedent that diamond shape patterns can be observed. This animal was entangled multiple times over a number of years. It is highly likely that this pattern was induced by a netting structure of some kind. Below the circle in this image there are other lines reminiscent of the current case.



Images by Jooke Robbins, Provincetown Center for Coastal Studies (PCCS). Injuries produced by tiger shark attack on live humpback whale calf, Hawaii. March 21, 2002. HIHWNMS/CCS, NOAA permit number 932-1489.

This humpback whale calf was alive and free-swimming throughout 2.5 hours of observation. It was a neonate that had been separated or abandoned and was estimated to be within a day or two of birth. The calf was essentially eaten alive over that period of time and no useful parts of the carcass were ever recovered. So, all of the injuries were quite fresh and no physical sampling was possible. The shark made many attacks, from different directions (including by rolling itself over the dorsal peduncle and back) and the calf was continuously swimming and struggling against them. The event was already well underway when the observers arrived, but this took place in Hawaii (on a neonate) and in situ entanglements there are rare (nor was there any direct evidence of such in this case). Ms Robbins therefore feels, on the basis of this experience that all of the defects observed in the current case could be shark induced. Scott Landry of PCCS concurs).







Assessment of photographs of dead calf in relation to possible entanglement interaction – Amy Knowlton, New England Aquarium – Jan 30th 2006

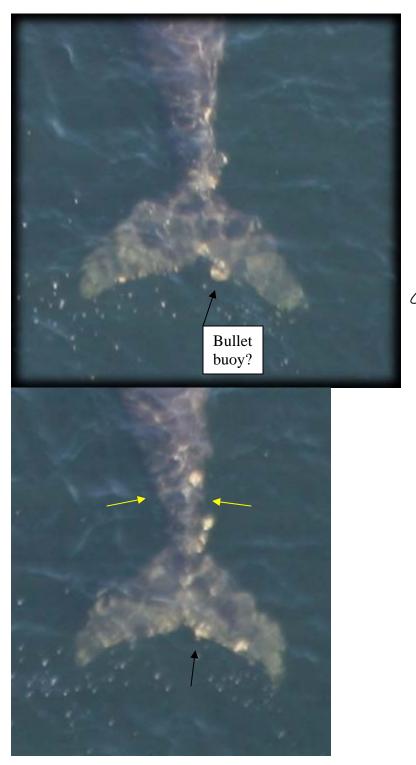
The New England Aquarium has continued to evaluate images of the dead right whale calf found on 1/22/06 off the Florida coast. We have also evaluated images of this calf when still alive from December 30, 2005 and January 8, 2006. Based on our continuing evaluation, we believe this animal suffered from an entanglement interaction based on the following evidence (which is in addition to that presented by Philip Hamilton and included in Moore's draft report from 1/26/06). We also agree that there was shark interaction as well but do not feel that it was the sole cause of all the scars/lesions observed on the living and dead calf images. Though we recognize the photo quality is poor for assessing this interaction thoroughly, we do believe that enough evidence exists to indicate an entanglement interaction. We here provide drawings/arrows of our interpretations next to or within the images:

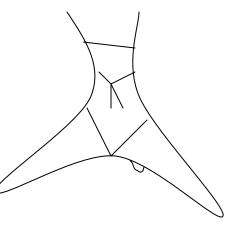
- 1) Based on several images from January 8, 2006, there appears to be a peduncle wrap forward of the insertion area. The extent of this line extends from at least halfway down the left side of the peduncle to halfway down the right side of the peduncle. We cannot envision that this wrapping, straight-line lesion could have been incurred by a shark. Although it did not appear to go all the way around and dig in to the ventral peduncle, this could be because of the way it was being pulled by drag.
- 2) There appears to be line trailing back along the middle of the dorsal peduncle which forms a vee into a knot with two trailing bitter ends. In this same image, there appears to be a white object appearing from underneath the trailing edge of the right ventral fluke. It shows up in two sequential images and may be a bullet type buoy. This is difficult to confirm but if this was there, it could help explain the pattern of the line wrap described above, ie the drag of that buoy from underneath would have prevented the line wrap from embedding ventrally as it did dorsally.
- 3) There are two white line scars extending forward from the fluke notch for a short distance. If we interpret the direction of these lines, it would suggest that the gear would have extended over the leading edge of the flukes posterior to the insertion. In reviewing images of the carcass on the beach, there is indication of abrasion at the questionable area of the right leading edge. Also, on the ventral flukes, there appears to be some indication of similar type of scarring.
- 4) On the floating carcass images taken aerially, the pattern aft of the main peduncle wrap is of parallel lines that cross perpendicularly as well. Forward of the main ped wrap are lines that are irregular and not quite parallel (probably shark teeth marks).
- 5) Data indicates that sightings of right whales have occurred within 3 miles of the whiting gillnet gear on a couple of occasions already. Clearly the risk is there throughout the calving season from this new emerging fishery.

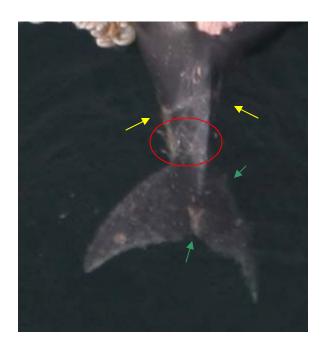
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16-Jan-06 30 26.0 x 081 19.4 No Sightings south of 30 31.0)
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30 27.0 and 81 19.0	
(at least 2 sets of	
24-Jan-06 1056 gear) No sightings south of 30 37.0	

Data From Chris Slay
Data from Andy Garrett
(FWC)

Data from EWS (NEAq)



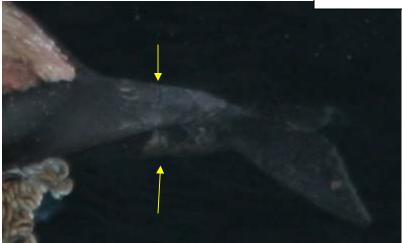




Criss-cross pattern in red circle – lines are parallel and perpendicular vs. striations forward of ped wrap which are irregular and not parallel

Line at yellow arrows extends around to right side (can be seen in Jan 8th aerials too)

Line at green arrows is very faint – should reassess dorsal fluke images from beach and see if they can be detected. Some indication of ventral impressions originating from notch. Not sure if cyamid filled vee extending from notch is related to this faint line or not.



APPENDIX EgNEFL0603 Page 34 of 34

From: "Skomal, Gregory (FWE)" < Gregory. Skomal@state.ma.us>

Date: Fri, 27 Jan 2006 09:46:25 -0500 To: "Michael Moore" <mmoore@whoi.edu>

Subject: RE: Need a consult

Hi Michael:

I've examined the report and the images of the dead right whale and do not believe that all the defects on the left peduncle are shark inflicted. The patterns, particularly the diamond shaped lesions (pg. 12), are not consistent with shark jaw morphology or my observations of shark bites. I concur that post-mortem lesions on the right peduncle are shark inflicted, and I am particularly intrigued by the two premortem predation bites. Is the latter commonly seen on right whale calves? At 22 cm across, these wounds were inflicted by a relatively small

With regard to the dorsal defect, the long slices seem too clean to be caused by a shark, but the ragged edges in the caudal and cranial regions could have been inflicted by a large shark. Stations B, D, and E give the appearance of tooth lacerations, but it would have had to been a very large animal, like a white shark, which is known to scavenge whale carcasses. The long smooth slices are not characteristic of shark bites and more closely resemble the clean slice of a boat propeller.

Please don't hesitate to contact me if you need me to elaborate on any of these opinions. I hope this is helpful.

Best,

Greq

----Original Message----

From: Michael Moore [mailto:mmoore@whoi.edu]

Sent: Thu 1/26/2006 4:48 PM To: Skomal, Gregory (FWE)

Cc:

Subject: Need a consult

 ${\tt Greg}$ - if you have time could you look at the images in this report. I assumed that the left peduncle lesions were entanglement related, but a colleague has suggested they might all be shark in origin. See ftp://ftp.whoi.edu/pub/users/mmoore_dropbox for the report. please dont circulate as it is still a draft. The interretation of the lesions is critical in terms of whether a fishery is implicated.

Michael Moore Biology Department Mailstop 50 Woods Hole Oceanographic Institution Woods Hole MA 02543 USA 508 289 3228 t 508 457 2089 f

www.whoi.edu/people/mmoore

1 of 1 2/1/2006 8:57 AM

SUMMARY OF FINDINGS: EGNEFL 0603

Primary Pathology Case Administrators:

David S. Rotstein, DVM, MPVM, DACVP University of Tennessee/NOAA-NMFS

Lisa L. Farina, DVM, DACVP University of Florida

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2.Appendix I: Integrated Pathology Report	7-35

Summary

<u>History</u>

A right whale calf (mother #1802) was sighted off the coast of Florida on 22nd of January. It was sighted alive on 12/30/2005, 1/8/2006, and 1/11/2006. Shark bites were evident on 12/30/2005. In addition to these initial lesions, other integument changes were observed on 1/8/2006. On 1/11/2006, there were no additional lesions observed.

Gross Findings

Externally, there was a dorsal disruption of epidermis and peduncle lesions. The large dorsal defect was approximately 2.4 m X 1.2 m and had associated extrusion of intestinal loops. Peduncle lesions ranged from multiple shallow to deep epidermal defects in the peduncle that formed semi-circles, linear, v-shaped, and diamond patterns. In addition, large numbers of Cyamids were observed in the peduncle region including in wound regions. Significant internal (post-cranial) findings were not observed.

Based upon these findings, the following gross diagnoses were generated:

A.Dorsal Defect: Regionally extensive epidermal defect with cranial and caudal irregular and roughened edges and segmental smooth edges without associated subdermal edema or hemorrhage.

B.Peduncle:

- 1.Regionally extensive epidermal and dermal indentation and penetration with overall pattern formation of semi-circles, linear, vees, and diamond with localized tissue reaction.
- 2. Multiple semi-circular cutaneous lesions with localized tissue reaction.
- 3. Cutaneous Cyamidiasis, severe.

Epidermal lesions in the peduncle were separated in regards to shape (diamond, vee, straight line) and pattern (regularity of pattern formation, irregularity or inconsistent pattern formation), In the case of the peduncle lesions, shapes that occurred in regular patterns included diamond, vee, and straight lines, and the shape that appeared with an irregular pattern were semi-circular.

Histologic Assessment

Sampling. Skin sections from the dorsal defect and from the peduncle were collected at "stations". Sections were submitted to two institutions, University of Florida and University of Tennessee for histologic evaluation. A listing of tissues, site of collection, size, and description are indicated in Table 1 reflecting samples received at the University of Tennessee. Corresponding gross photos of the integument may be viewed in Appendix 1. In addition to skin sections, internal organs (post-cranial) were also submitted and examined.

Table 1. Description of integument obtained at stations. This reflects samples received at University of Tennessee.

Integument Station ID	Size (cm)	Comment
A-Dorsal Lac†	4.5 X 4.0 X 0.7	White to yellow and rubbery
B-Dorsal Lac	7 X 4 X 0.7; epith*-1.5	Ragged bottom
C-Dorsal Lac	8 X 4.5 X 0.7; epith-1.5	Ragged bottom
D-Dorsal Lac	12 X 4.2 X 1.0; epith-1.6	-
E-Dorsal Lac	10.5 X 3.9 X 2.0; epith-1.5	Two superficial defects-round (0.5 cm diam) & wedge (1 X 0.5 x 0.2)
H-Peduncle	3 X 4 X 1	Epithelial wedge; whitening of epithelium; dermis gray
H2-Peduncle	3.5 X 4.9 X 1.0; epith 1.1	Central 0.5cm cleft; white at base; gray 1 X 1.5cm flask- shaped subjacent to epithelium
H3-Peduncle	4.6 X 6 X 0.9; epith 1.5	0.4cm epith cleft; white streaks in epith; 1.5 X 1cm gray wedge subjacent to epith
I-Dorsal Lac	2.5 X 3 X 1.0	Epith & dermis separated; dermis-gray
L-Peduncle	11.5 X 3.5 X 1.0; epith 1.3	Approx. 9.0 cm region of epith loss; whitening of epithelium
N-Dorsal Lac	8 X 6.5 X 1.5; epith 1.5	At edge, 1.5 X 4.5 X 0.6 cm region of dermal lightening
O-Dorsal Lac	10.5 X 8.1 x 1.5; epith 1.6	Focal 1.0cm cleft
P-Peduncle	5.2 X 4.2 X 1.0; epith 1.0	There is focal 1.0cm cleft
Q-Peduncle	2.4 X 4 X 1.0	Epithelial cleft; whitening of epithelium
R-Peduncle	5 X 4 X 3	Overriding epithelium; whitening

†Please note that LAC, standing for laceration is terminology used for received samples and is synonymous with the term "defect" utilized in the gross report, microscopic findings, and discussion with the histology report *epith= epithelial thickness

There was minimal inflammation within the dermis of sections from the dorsal defect and minimal edema. Hemorrhage was not observed. All peduncle lesions were characterized by orderly formation of granulation tissue and all had focal to regionally extensive epithelial lesions (ulceration). Granulation tissue formation is characterized by edema with inflammatory cells and loose fibroblasts transitioning to more organized densely packed fibroblasts interspersed with vascular channels. In one section (I), inflammation was more pronounced and the dermis contained a granuloma. In another section (L), there were superficial intradermal, herniated nests of epithelial cells (suspected traumatic implantation). Epithelial hyperplasia was present in all peduncle sections examined.

Sample Evaluation

At the end of the examination, pathologists from both institutions had a full review of histologic sections. In addition, a review of the integument lesions was conducted by a pathologist from the Armed Forces Institute of Pathology (AFIP) (M.Fleetwood). Gross photos and integument lesions (histologic sections) were also reviewed by a forensics pathologist from the medical examiner's office for Knox County (Mileusnic), and gross photos were also examined by a forensic anthropologist and engineer with boattrauma experience (T. Kress, A. Korman). Finally, the summary and final report were reviewed by two additional veterinary pathologists (D. Miller, T. Zabka).

Comment

Overall: The cause of death remains open in this case. Organs were autolyzed, which limited histologic assessment; however, there was no appreciable evidence of systemic disease in the examined sections. While there were significant peduncle lesions, the effect upon this calf (e.g. morbidity/mortality) is speculative. Gastric contents were absent, indicating that this animal did not nurse recently. Such a change could be an indication of debilitation, but other causes cannot be excluded. If this animal did not

eat, then metabolic and electrolyte derangements could have occurred and lead to death. Unfortunately, samples were not available (due to autolysis) for clinical chemistry analysis to rule out a metabolic or electrolyte imbalance.

There were two regions (one on the dorsum and one on the peduncle) of integument with evident gross changes. The dorsal defect is unique in its location and appearance. A section of intestine protrudes from this site suggesting postmortem intrabdominal and intra-viscous gas production and expulsion. In such an occurrence, hemorrhage and inflammation would not be expected in affected tissues, and in this case, there was negligible associated inflammation and edema. Also, post-mortem gaseous expulsive events are typically abdominal (ventral), which was not the case in this right whale. With these slightly contradictory findings, possible scenarios include that the animal was listing in the water such that the path of least resistance in the case of increased pressure was dorsal, or that there was an initial penetrating event leading to a region of weakened epithelium, separation of the integument, and eventual expulsion of an intestinal segment. Although hemorrhage and significant inflammation were not observed at this site, there was very mild edema and scattered inflammatory cells suggesting antemortem (or at least perimortem) development. The underlying causes for these presumed antemortem changes were not evident, but could could have resulted from localized vascular leakage or systemic vascular leakage. These latter changes were not interpreted as having contributed to the development of the dorsal wound. Overall, bloat was considered the cause for the skin defect and intestinal herniation by all parties involved in the pathologic evaluation. A final comment is that two reviewers (Kress and Korman) raised concern about two sites of the dorsal wound as possible traumatic injury. Regardless, changes in the dorsal defect were interpreted as post-mortem event.

Potential scenarios for the development of the peduncle lesions include predator, anthropogenic insults, or mixed predator and anthropogenic (e.g. net entanglement) causes. As described above, lesions varied in the peduncle from irregular semi-circle to regular linear, vees, and diamonds. Based on these changes, it is the opinion of the two institutions that examined the gross wounds from photos, that the lesions likely reflect mixed causes. Specifically, anthropogenic causes, such as net entanglement may have caused the linear lesions, diamonds, and vees; whereas, predator (shark) attack (i.e. shark bites) may have resulted in the irregular semi-circular lesions. Furthermore, even in post-mortem predation, linear, regular wounds are not a common finding. Histologically, these wounds either had the greater inflammation or presence of intradermal-epithelial nests that suggest implantation. The semi-circular wounds were grossly and from aerial surveys sites most consistent with shark bites. This said, all lesions that involved defects extending into the dermis from the peduncle had evidence of granulation (healing). Wound repair in some sections did vary slightly suggesting that these wounds may have occurred at differing time points; however, local and systemic effects (e.g. mobility at site, size of lesion, infection, systemic disease), may affect healing rates.

In summary, this right whale calf had a striking and unique dorsal wound of an uncertain cause, but most likely due to a post-mortem event that in some degree related to bloat. The calf also had multiple peduncle lesions that most likely are due to a combination of predator and human sources. The exact cause of death remains open.

David S. Rotstein, DVM, MPVM, DACVP

Appendix 1: Final Necropsy Report

CETACEAN NECROPSY REPORT: INTEGRATED REPORT-University of Florida & University of Tennessee/NMFS

Field ID:EGNEFL0603

Accession Number: 06-004C Species: Eubalaena glacialis

Date: 1/22/2006

Location: FL (30 14.76N, 81 03.2 W)

Sex: F

TL: 560.0 cm

GROSS REPORT

REPORTING INSTITUTION: University of Tennessee

PDF file received.

ANCILLARY FINDINGS

Received 25 Jan 2006 is one bag containing unfixed, individually bagged adrenal gland, liver, lung, and ovary. Disposition of tissues is as follows:

Virology:

- 1. Isolation: Oklahoma Animal Disease Diagnostic Laboratory (OADDL) and National Veterinary Service Laboratories (NVSL).
- 2.PCR-Morbillivirus and Herpesvirus: OADDL
- 3. Tissues Submitted: Adrenal Gland, Liver, Lung, Lymph Node, Ovary
- 4. Tissues Frozen: Adrenal Gland, Liver, Lung, Ovary
- 5.Results:1 Feb 2006-Herpes and Morbillivirus PCR-NEG

Bacteriology:

Ovary: Brucella, Mystic Aquarium

TISSUES/SAMPLES RECEIVED

REPORTING INSTITUTION: UNIVERSITY OF TENNESSEE

Received 27 January 2006 are three bags labeled as "EGNEFL0603 Histo". For the purpose of this gross description, bags were randomly numbered 1-3. **Bag 1** contains 6 sections of integument which are labeled as "Station" followed by "A", "B", "C", "D", "E", and "I" which are described in the table below. **Bag two** contains 31 tagged tissues including "Station Q shark bite" and "lesion R". Labeled tissues are measured in **cm** (parentheses) with representative sections placed into cassettes as follows: 8-aora (3.1 X 0.9 X 0.6), adrenal gland (2 X 1 X 0.5), 1st stomach (3.5 x 2.5 X 1.0), lung (2 X 1 X 0.5); 9-pulmonary artery (2 X 0.5 X 1), kidney (6 X 2.5 X 1.0); 10-right papillary muscle, heart (2 X 1.3 X 0.3), bile duct (5 X 1.5 X 1); 11-adrenal gland (?) (2 X 1.5 X 0.9), distal colon (3 X 2 X 0.5); 12-uterus (3 X 1.5 X 0.4), 2nd stomach (3 X 1 X 1), liver (5 X 2 X 0.6); 13-3rd stomach (4.5 X 2 X 1), right atrium (3 X 2.0 X 0.5); 14-thymus (7 X 2.5 X 1.0), right lung (4 X 2 X 0.3); 15-trachea (3.5 X 1.0 X 0.5), diaphragm (6 X 3 X 0.5), ovary (3.5 X 1.0 X 1.0); 16-vagina (3 X 1 X 1), mediastinal lymph node (4.5 X 2.0 X 0.6); 17-cervix (2.5 X 3 X 1), right ventricle (3.5 X 1 X 1); 18-urinary bladder (4 X 1.5 X 0.5), left ventricle (3 x 2 X 0.5); 19-esophagus (6 X 2 X 0.5); 20-distal esophagus (3.5 X 1 X 1; lumen-2.0cm). **Bag three** contains 8 tagged and labeled tissues as well as 7 integument sections labeled as "Station" followed by "H", "H2", "H3", "O", "N", "P", and "L". These are described below. Labeled tissues are measured in **cm** (parentheses) with representative sections placed into cassettes as follows: 1-tongue anterior (3 X 4.2 X 0.5); 2-tongue mid (5 X 3 X 0.7); 3-tongue posterior (3.5 X 2.6 X 0.6); 4-mid ventral muscle edema, right thorax (2.5 X 1.5 X 1.0), right thyroid gland (8 X 1.5 X 0.5); 5-mid lat muscle edema (2 X 4 X 1.5); 6-vent midline edema "F" (4.5 X2 X 1); 7-ven midline edema "D" (4.5 X2 X 1.0).

Table 1. Description of integument obtained at stations.

Integument Station ID	Size (cm)	Photo¥	Comment	Cassette
A-Dorsal Lac†	4.5 X 4.0 X 0.7	Stellar F. Breel Loc	White to yellow and rubbery	21
B-Dorsal Lac	7 X 4 X 0.7; epith-1.5	Poly Hope	Ragged bottom	22, 23
C-Dorsal Lac	8 X 4.5 X 0.7; epith-1.5	John Mc	Ragged bottom	24
D-Dorsal Lac	12 X 4.2 X 1.0; epith-1.6	Specification of the second		25, 26
E-Dorsal Lac	10.5 X 3.9 X 2.0; epith-1.5	Age to	Two superficial defects-round (0.5 cm diam) & wedge (1 X 0.5 x 0.2)	27, 28
H-Peduncle	3 X 4 X 1	Station 19	Epithelial wedge; whitening of epithelium; dermis gray	37-39
H2-Peduncle	3.5 X 4.9 X 1.0; epith 1.1	Station 112	Central 0.5cm cleft; white at base; gray 1 X 1.5cm flask-shaped subjacent to epithelium	35, 36
H3-Peduncle	4.6 X 6 X 0.9; epith 1.5	Shiften H3	0.4cm epith cleft; white streaks in epith; 1.5 X 1cm gray wedge subjacent to epith	33, 34
I-Dorsal Lac	2.5 X 3 X 1.0	Shtin I	Epith & dermis separated; dermis-gray	31

L-Peduncle	11.5 X 3.5 X 1.0; epith 1.3	Approx. 9.0 cm region of epith loss; whitening of epithelium	of 42, 43
N-Peduncle	8 X 6.5 X 1.5; epith 1.5	At edge, 1.5 X 4.5 X 0.6 cm region of dermal lightening	46-48
O-Dorsal Lac	10.5 X 8.1 x 1.5; epith 1.6	Focal 1.0cm cleft	29, 30
P-Peduncle	5.2 X 4.2 X 1.0; epith 1.0	There is focal 1.0cm cleft	32
Q-Peduncle	2.4 X 4 X 1.0	Epithelial cleft; whitening of epithelium	40, 41
R-Peduncle	5 X 4 X 3	Overriding epithelium; whitening	44,45

†Please note that LAC, standing for laceration is terminology used for received samples and is synonymous with the term "defect" utilized in the gross report, microscopic findings, and discussion with the histology report. ¥Photos by M. Shane Cummings, UT

MICROSCOPIC FINDINGS

REPORTING INSTITUTION: UNIVERSITY OF TENNESSEE

NOTE BENE: There is mild to moderate autolysis in most tissues characterized by post-mortem bacteria (cadaver bacilli) and post-mortem gaseous distention of tissues. This statement is considered universal for tissues examined and will not be repeated for each slide description, allowing focus on observed changes.

Slide 1 (UF Slide 26)

Tongue, Anterior: Three sections are examined. Within the submucosa, there is mild, focal erythrocyte extravasation separating collagen fibrils (Figure 1) (on Slide 1 only).

Slide 2 (UF Slide 36)

Tongue, Mid: Three sections are examined. Two sections are examined. Occasionally, within the cytoplasm of stratified squamous epithelium, there are variably-sized (4 to 10 micron), irregular oval to round, hyaline globules (Figure 2). There is mild vascular congestion.

Slide 3 (UF Slide 42)

Tongue, Posterior: Three sections are examined. There is mild, vascular congestion. Occasionally, within the cytoplasm of stratified squamous epithelium, there are variably-sized (4 to 10 micron), irregular oval to round, hyaline globules.

Slide 4 (UF Slide 24)

Mid-Ventral Muscle Edema, Thorax, Right: Two sections are examined. There is multifocal separation of myofibers and collagen fibrils by pale, pink, finely granular substance.

Thymus (labeled as right thyroid gland): Cellularity is moderate. The distinction between cortex and medulla is not apparent. Cellular morphology is indistinct (autolysis).

Slide 5

Mid-Lateral Muscle Edema: Two sections are examined. There is multifocal separation of myofibers and collagen fibrils by pale, pink, finely granular substance.

Slide 6 (UF Slide 27)

Ventral Midline Edema "F": Three sections are examined. There is multifocal separation of myofibers and collagen fibrils by pale, pink, finely granular substance.

Slide 7 (UF Slide 45)

Ventral Midline Edema "D": Three sections are examined. There is multifocal separation of myofibers and collagen fibrils by pale, pink, finely granular substance.

Slide 8

Lung (UF Slide 37): NSF.

Adrenal Gland (UF Slides 25 & 28): Tissue on Slide 8 consists of nerve bundles. Tissue in UF Slides 25 & 38 is markedly autolyzed adrenal gland, with extensive post-mortem bacterial overgrowth.

1st Stomach (UF Slide 33): No significant histologic findings (NSF).

Slide 9

Pulmonary Artery (UF Slide 35): Occasionally, there is separation of collagen fibers by pale spaces (Slide 9). Rarely, there are foamy macrophages within these regions (Figure 3). Tissue on UF Slide 35 contains extensive post-mortem bacterial overgrowth and few clear, distinct vacuoles within the tissue.

Kidney: NSF. Glomeruli are not distinct. There is a single organism that is approximately 500 microns in width with a chitinous cuticle, striated skeletal muscle, digestive tract, and reproductive organs (suspect Cyamid). There is not associated inflammation—suspected artifactual introduction).

Slide 10

Right Papillary Muscle, Heart (UF Slide 36): NSF.

Bile Duct (UF Slide 34): NSF.

Slide 11

Adrenal Gland (UF Slides 25 & 28): The examined section is nerve with no significant histologic findings. Tissue in UF Slides 25 & 38 is markedly autolyzed adrenal gland, with extensive post-mortem bacterial overgrowth.

Distal Colon (UF Slide 32): NSF.

Slide 12 2nd Stomach (UF Slide 30): NSF.

Uterus (UF Slide 41): Three sections are examined and consist of myometrium with NSF.

Slide 13

3rd Stomach (UF Slide 44): NSF.

Right Atrium (UF Slide 39): NSF.

Slide 14

Thymus (UF Slide 38): There is moderate cellularity, but cellular morphology is indistinct save for a small region where there are fairly distinct small lymphocytes. The distinction between cortex and medulla is poor.

Right Lung (UF Slide 32): Alveolar lumina contain granular to globular eosinophilic substance and occasional foamy macrophages (Figure 4).

<u>Slide 15</u>

Diaphragm (UF Slide 30): NSF.

Trachea (UF Slide 46): NSF.

Ovary (UF Slide 42): Primary follicles are present, but morphology is indistinct.

Slide 16

Vagina (UF Slide 31): NSF.

Mediastinal Lymph Node (UF Slide 40): The section consists of lobules and represents thymus. Findings are similar to Slide 14.

Slide 17

Cervix (UF Slide 31): NSF.

Right Ventricle: NSF.

Slide 18

Urinary Bladder (UF Slide 33): There is a focal aggregate of mineral deep within the muscularis (UF Slide 33).

Left Ventricle (UF Slide 29): NSF.

Slide 19

Esophagus (UF Slide 35): Mucosal epithelium is artifactually sloughed.

Slide 20

Distal Esophagus (UF Slide 43): Mucosal epithelium is artifactually sloughed.

Dorsal Defect:

Slide 21 (UF Slide 1)

Station A, Dorsal Defect: Four sections are examined. Sections consist of collagen bundles and adipose. NSF.

Slides 22-23 (UF Slide 2)

Station B, Dorsal Defect: Four sections are examined. There is superficial intracellular expansion by eosinophilic fluid. There is mild separation of collagen fibers by clear spaces (edema) with rare macrophages associated within the superficial dermis. Within the dermis, there is occasional adipocyte disruption characterized by loss of thin, adipocyte cell membranes (Figure 5). Inflammation and hemorrhage are not associated.

Slide 24 UF Slide 3)

Station C, Dorsal Defect: Four sections are examined. There is superficial intracellular expansion by eosinophilic fluid. Within the dermis, there is occasional adipocyte disruption characterized by loss of thin, adipocyte cell membranes. Inflammation and hemorrhage are not associated. There is mild dermal edema with a few interspersed macrophages observed.

Slides 25-26 (UF Slide 4)

Station D, Dorsal Defect: There is superficial intracellular expansion by eosinophilic fluid. Within the dermis, there is occasional adipocyte disruption characterized by loss of thin, adipocyte cell membranes. Inflammation and hemorrhage are not associated.

Slide 28 (UF Slide 5)

Station E, Dorsal Defect: Three sections are examined. There is superficial intracellular expansion by eosinophilic fluid. Within the dermis, there is occasional adipocyte disruption characterized by loss of thin, adipocyte cell membranes. Inflammation and hemorrhage are not associated.

Slides 29-30 (UF Slides 15-17):

Station O: Six sections are examined. Approximately 5 cell layers deep into the stratum externum, there is a laminar band of epithelial cells separated by lakes of eosinophilic, proteinaceous fluid (Figure 6). There is separation of the stratum basale from the papillary dermis. The papillary dermis is expanded is pale spaces and within the fibroadipose, there are occasional foamy macrophages. Adipocytes are occasionally expanded with loss of septa and a granular eosinophilic substance filling the spaces.

Slides 46-48 (UF Slide 28)

Station N: Four sections are examined. Findings are similar to Slides 29-30 (Station O).

Right Peduncle

Slide 31 (UF Slide 14)

Station I: Four sections are examined. Changes are observed within the epithelium and dermis. There is irregularity of the surface epithelium and multifocal to confluent swelling of epithelial cell cytoplasm (hydropic degeneration). Nuclei of affected cells are shrunken. Multifocally, within the basal layer and stratum intermedium, there is loss of pigment within epithelial cells. The epidermis at the margins of the defect is hyperplastic. There is separation of the stratum basale from the papillary dermis. Within the papillary dermis and reticular dermis, there is separation of collagen by pale spaces (edema) with numerous interspersed lymphocytes, plasma cells, macrophages and fewer neutrophils (Figure 7) (neutrophils on UF Slide 14). In one region (in Slide 31 only), there is a well-circumscribed dermal nodule with surrounding layers of epithelioid macrophages, and a core of degenerate neutrophils and macrophages flanking, lacy eosinophilic material and a few Gram-positive bacterial rods (Figure 8). Acidfast (Mycobacteria), Giemsa (Protozoa), PAS(Fungus) and GMS (Fungus) stains were negative. Fibrin thrombi are present within vessels of the exposed papillary dermis, and there is a single vessel which is filled with fibrin with segmental loss of the endothelial wall and surrounding mononuclear cells (Figure 9). Extending from the superficial dermis to the deeper dermis, there are fibroblasts with tinctorial change from lighter to darker pink in deeper layers (increased collagen production) and decreased cellularity replacing fibroadipose (Figure 10).

Slide 32 (UF Slide 18)

Station P: Two sections are examined. Findings in the dermis are similar to Slide 31 with less inflammation observed. In addition, within the superficial dermis, there is a sprinkling of perivascular neutrophils. Hydropic change in the epithelium is less prominent than Slide 31. Pigment is decreased to absent within the stratum basale and intermedium.

Slides 33-34 (UF Slide 11)

Station H3: Four sections are examined. In two sections (slide 34 and UF Slide 11), there is a focal epithelial defect. Vessels within the papillary dermis at the margins of this defect exhibit marked perivascular collagen accumulation and endothelial plumpness (reactivity) (Figure 11). Lining epithelial cells exhibit mild to moderate hydropic change and there is decreased pigment within epithelial cells. The epidermis at the margins of the defect is hyperplastic. In the subjacent dermis, there is an irregular vertically oriented expansion of the dermis by clear spaces (edema) with interspersed plump fibroblasts replacing fibroadipose. Macrophages and fewer plasma cells and lymphocytes are sprinkled within this affected region as are small islands of intact collagen.

Slides 35-36 (UF Slides 8-10)

Station H2: Seven sections are examined. In two sections (slide 36 and UF Slide 8), there is a focal epithelial defect. Vessels within the papillary dermis at the margins of this defect exhibit mild to moderate perivascular collagen accumulation and endothelial plumpness (reactivity). Lining epithelial cells exhibit mild to moderate hydropic change and there is decreased pigment within epithelial cells. The epidermis at the margins of the defect is hyperplastic. In the subjacent dermis, there is an expansion of the dermis by clear spaces (edema) with interspersed, loosely arranged plump fibroblasts (Figure 12). Macrophages are sprinkled within this affected region as are small islands of intact collagen. Plasma cells and lymphocytes are occasionally observed. Vascular channels are markedly ectatic. The epithelial surface is irregular.

Slides 37-39 (UF Slides 6-7)

Station H: Six sections are examined. There is an epithelial defect extending to the basal layer. In this region, there is an abrupt transition from epithelial cells with black, granular pigment to a lack of pigment (leukoderma) (Figure 13). The epidermis at the margins of the defect is hyperplastic. There is multifocal superficial dermal edema with few interspersed macrophages and loosely arranged, fibroblasts. In one section (Slide 37), there is perivascular expansion by clear spaces (edema) and within this expanded region, there are variably-sized, 4 to 6 micron, irregular round, amphophilic structures with either a solid core, or a lightly vacuolated core (Figure 14) (Slide 37 only). Inflammation is focal and mild, consisting of lymphocytes and plasma cells, typically oriented perivascularly. In one section (Slide 39), regions of fibrosis are characterized by increased collagen and decreased cellularity and nuclear size.

Slides 40-41 (UF Slides 19-21)

Station Q: Five sections are examined. Superficial layers of the stratum externum are irregular. Approximately 5 to 8 cells layers subjacent to the surface, there are single to multilocular, irregular round to oval to dacryoid clear spaces separating epithelial cells. These spaces occasionally contain pale pink, proteinaceous fluid. In the epithelium at the site of grossly observed separation at the epithelial-dermal interface, there is marked cytoplasmic swelling of epithelial cells (hydropic change) (Figure 15). Pigment is absent from these affected epithelial cells. There appears to be regional epidermal hyperplasia. There is regionally extensive dermal edema with interspersed macrophages, fewer lymphocytes and plasma cells. Fibroblasts are increased replacing fibroadipose and forming loosely arranged bundles.

Slides 42-43 (UF Slides 12-13)

Station L: Six sections are examined. Superficial layers of the stratum externum are irregular and there is regionally extensive epithelial loss. There is multifocal cytoplasmic swelling of epithelial cells (hydropic change). Pigment is absent from these affected epithelial cells. There is mild dermal edema and regionally extensive replacement of fibroadipose by bundles of fibrocytes separated by abundant collagen. Lymphocytes, plasma cells, and macrophages are present occasionally in small aggregates. In one section (Slide 43), at the site of extensive epithelial loss, there are small islands of epithelium within the superficial dermis surrounded by small numbers of lymphocytes and plasma cells.

Slides 44-45 (UF Slides 22-23)

Lesion R: Five sections are examined. Superficial layers of the stratum externum are irregular and there is regionally extensive epithelial loss. There is multifocal cytoplasmic swelling of epithelial cells (hydropic change). Pigment is absent from these affected epithelial cells. The epidermis at the margins of the defect is hyperplastic. Fibroadipose is replaced in two regions, one subjacent to the stratum basale and the other separated by a layer of intact, unaffected fibroadipose. In the former, there is abundant collagen and decreased cellularity. In the latter, there is moderate edema, loosely arranged fibroblasts, and scattered macrophages with fewer lymphocytes and plasma cells.

MORPHOLOGIC DIAGNOSES

Respiratory System:

Lung: Alveolar histiocytosis, multifocal to confluent, mild, presumptive.

Integumentary System:

- A. Dorsal Defect (Stations, A-E, N, O):
 - 1. Adipocyte membrane separation, multifocal (post-mortem artifact).
 - 2. Station O: Histiocytic dermatitis, multifocal, minimal.
 - 3.Edema, minimal.

B. Peduncle:

1. General:

Superficial epidermal erosion (Ectoparasite related, suspect) with associated superficial epithelial cell hydropic degeneration.

2. Stations:

a) I:

- 1) Transmural epithelial cleft formation with subjacent fibroplasia and lymphoplasmacytic, neutrophilic and histiocytic dermatitis, focal granulomatous dermatitis, mild to moderate dermal edema, superficial intravascular fibrin thrombi, and epithelial hyperplasia.
- 2) Pigment loss (leukoderma).

b) H, H2, H3:

- 1) Transmural epithelial cleft formation with subjacent fibroplasia, mild lymphoplasmacytic and histiocytic dermatitis, mild to moderate dermal edema, perivascular collagen accumulation and endothelial hypertrophy.
- 2) Pigment loss (leukoderma).
- 3) Perivascular structures, focal, dermal (Station H) (SEE COMMENT).

c) L:

- 1) Regionally extensive cutaneous ulceration with subjacent collagen deposition, multifocal, mild lymphoplasmacytic and histiocytic dermatitis, superficial intradermal epithelial nests, mild to moderate dermal edema and intravascular fibrin thrombi.
- 2) Pigment loss (leukoderma).

d) P, Q:

- 1) Transmural epithelial cleft formation with subjacent fibroplasia and lymphoplasmacytic and histiocytic dermatitis, and mild to moderate dermal edema.
- 2) Pigment loss (leukoderma).

FINAL DIAGNOSES/INTERPRETATIVE SUMMARY

Peduncle:

Fibrosis (Wound Healing)
Epidermal erosion (Cutaneous cyamidiasis)

Overall Cause of Death: OPEN.

Gross findings indicate two major cutaneous insults, a large dorsal defect and peduncle lesions including regions of epithelial loss (suspect shark bite) and cutaneous indentations forming V-shapes, diamonds, and irregular lines. While gross examination of these changes *in toto* can lead to the generation of potential or conclusive causes, histologic examination of tissues can only provide information regarding timing of cutaneous lesions. It is not an exact determination, but rather can indicate:

- a. Whether all regions exhibit similar histologic changes.
- b. Whether differences indicate differences in duration of lesions.
- c. Potential etiologies such as infectious agents (bacteria, fungus, etc.).

The histologic feature in question based upon the initial gross findings (and subsequent histologic examination) is that of wound repair. It is important to point out that while the process of wound repair (both first and second intention) has certain inherent processes, it may be affected by systemic and local host factors including nutrition, metabolic status, circulatory status, hormones, infection, mechanical factors (motion), foreign bodies, and finally, size, location, and type of wound (Cotran et al., 1999). This is important to consider because all of these may delay healing and therefore influence healing times.

Thus, before describing lesions observed, it is important to understand the process of wound healing by first (apposing edges) and second (wounds with separated edges) intention. Regardless of first or second intention, wound healing involves an orderly process that includes: induction of acute inflammatory response, regeneration of parenchymal cells, parenchymal and connective tissue cells proliferation and migration, extracellular matrix protein production, connective tissue remodeling, and collagenization and acquisition of wound strength. In summary, inflammation is the first response, followed by fibroplasia, then tissue remodeling and scarring. The resulting scar, at least in human studies, is 10% of strength in the first week and by 12 weeks, reaches 70 to 80% of the tensile strength of unaffected skin (Cotran et al., 1999).

In terms of timing, inflammation generally lasts 3 days with fibroplasia beginning on the first day and continuing to day 10. Final wound contracture begins day 3 and continues to at least day 30. This time reference is based upon a human study (Clark, 1991). Again, this is for an uninfected wound. At the time of wound creation, blood and fibrin fill the space forming a clot. Within 24 hours, neutrophils accumulate and the stratum basale will become hyperplastic at the wound edges. Epithelial cells will migrate along the margin of the wound. By day 3, macrophages will accumulate, replacing neutrophils [in infected wounds, neutrophils persist]. Granulation tissue, which includes new vessels, extracellular matrix, and fibroblasts, will infiltrate the space. By day 5, granulation tissue fills the wound and vessels within the granulation bed are at their zenith. In week 2, collagen is increasingly deposited and fibroblasts continue to proliferate. Neutrophils, edema, and vessels have diminished to disappeared. By the end of the first month, there is an absence of inflammatory cells and the epithelium is intact. The preceding describes healing by first intention, that is, the wound edges are well apposed. In second intention wound healing, the inflammatory reaction is more intense and more long-lived, granulation tissues is present in large amounts, and, finally, closure involves contraction leading to an actual reduction in size to 5 to 10% of the original area. In scar formation, there is blanching of the skin. In the case of the cetaceans, when full wound closure is present, there is white streaking to confluent whitening of the skin. This corresponds to depigmentation or **leukoderma**. Leukoderma is not uncommon at sites of cutaneous trauma.

Dorsal Defect. Multiple sections of skin representing circumferential sections of the defect were examined. In some sections, there are few superficial dermal macrophages and mild edema. The exact cause of the dorsal defect was not determined, but the absence of a marked inflammatory response makes an initial infectious or traumatic cause for epidermal dehiscence unlikely. There were vacuoles in the fibroadipose which likely were due to gas production by bacteria post-mortem. This was most likely a post-mortem defect secondary to gas expansion and subsequent rupture of the body wall.

Peduncle. While histologic features in all samples were consistent with wound healing, there were some differences in the histologic appearance of tissue from different sites. As stated above, edema, inflammatory cells, fibrocytes, blood vessels, collagen, and maturation are components of wound healing. While most of the sampled sections either had completely intact epithelium, others had focal transmural linear and horizontal clefts with horizontal clefts at the dermal-epidermal junction, or had a more extensive loss of epithelium. In all samples from this region, there was irregularity of the superficial stratum externum with or without hydropic change in epithelial cells. This is likely the result of cyamids feeding upon the epithelium. Hydropic change was also observed in epithelial cells lining clefts or defects in the epithelium, indicating localized epithelial response.

General trends were observed. Sections from Stations I, P, and L had a similar histologic appearance. Histologic lesions in these sections included superficial to mid-dermal edema with moderate inflammation including lymphocytes, plasma cells, and macrophages and rarely granulomas (Station I) with loosely interspersed fibroblasts superficially, and increased organization of fibroblasts in deeper layers. Thus, these lesions are considered to be of a similar time frame. Sections from Station Q and R were also similar, though differing slightly in degree of edema and fibrosis. Finally, three sections were examined from Station H, H2, and H3. No sections exhibited evidence of complete collagenization with loss of inflammatory cells, fibrocytes, and vessels. Thus, all wound healing is active, but there are slight differences in the appearances of the wounds from different sites, which suggests these lesions are of slightly different ages and/or were differentially affected by local or systemic factors that can affect the rate of wound healing. Local factors include mobility at infection at the site. Although internal tissues exhibited a great degree of autolysis, there is not an indication of systemic disease. The influence of stress hormones cannot be determined. The exact cause of these peduncle wounds is not apparent with histologic examination, but all represent wounds that extended into the dermis.

Station I had a single small dermal granuloma which is considered clinically insignificant. There were a few Gram-positive bacilli in the granuloma, but these were similar to post-mortem bacteria, so the significance of the bacteria likely represents post-mortem overgrowth rather than an initiating cause. In addition, compared to other examined sections, inflammation was greatest in sections from this station. Within the superficial dermis in Station L, there were a few small discrete islands of epidermis. These could have been traumatically introduced into the dermis, although that is speculative.

Systemic Lesions. Autolysis was moderate to marked in all sections which, while it does hinder histologic assessment, does not completely prevent some observations, such as inflammation. There were no lesions to suggest significant systemic disease.

Cause of Death. The cause of death of this calf remains open. It is possible that whatever the source or sources of inciting damage to the peduncle led to localized compression of nerves leading to decreased mobility, the wounds led to sepsis (histologically not evident, although autolysis may have precluded identification of subtle lesions in some organs), or this cetacean was debilitated from the wounds and was not able to continue on with the mother leading to a lack of feeding, starvation, and subsequent electrolyte disturbances and metabolic dysfunction. There is no supportive evidence for any of these scenarios, and they are all speculative. Alternatively, death was not related.

Peduncle Wounds and Dorsal Defect-Gross. The cause of the peduncle wounds may be best determined by examination of gross photos. All/most reviewing parties agreed that a multifactorial cause is most likely. The semi-circular lesions are most consistent with shark-bite and the v-shaped and linear lesions are most consistent with as human interaction.

References

1.Cotran, R. S., V. Kumar, and T. Collins. 1999. Tissue repair: Cellular growth, fibrosis, and wound healing *In* Robbins Pathologic Basis of Disease. W. B. Saunders Co., Philadelphia. Pp. 89-111. 2.Clark, R. A. 1991. Physiology, Biochemistry, and Molecular Biology of the Skin. Goldsmith, LA (ed). Oxford Press, New York. P. 571.

FIGURES

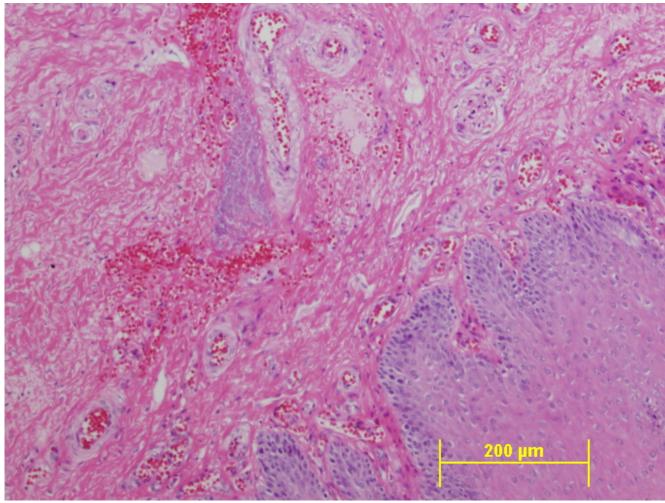


Figure 1. Anterior toungue. Small regions of erythrocyte extravasation. Vascular channels are congested.

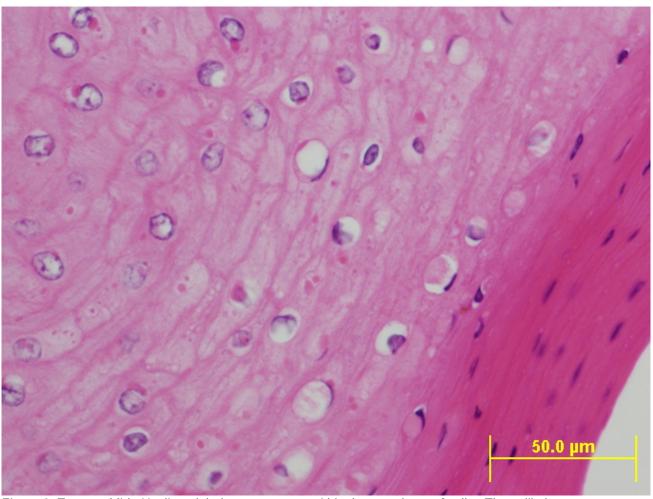


Figure 2. Tongue, Mid. Hyaline globules are present within the cytoplasm of cells. These likely represent keratinaceous product.

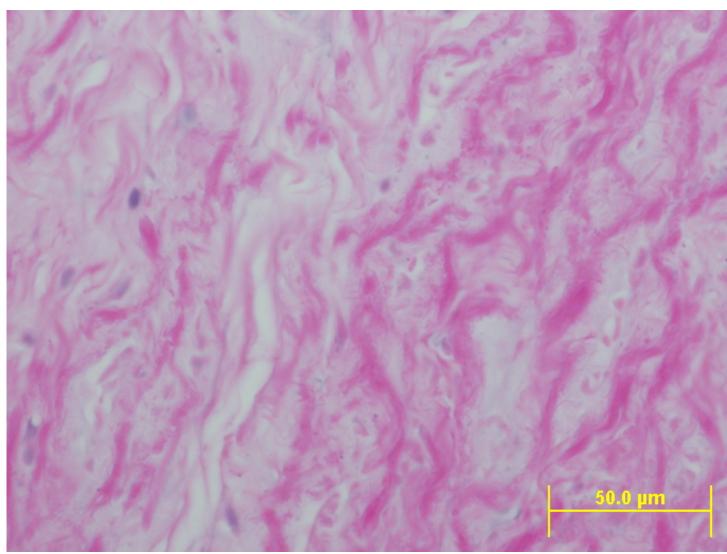


Figure 3. There is separation of connective tissue fibers of the pulmonary artery (edema).

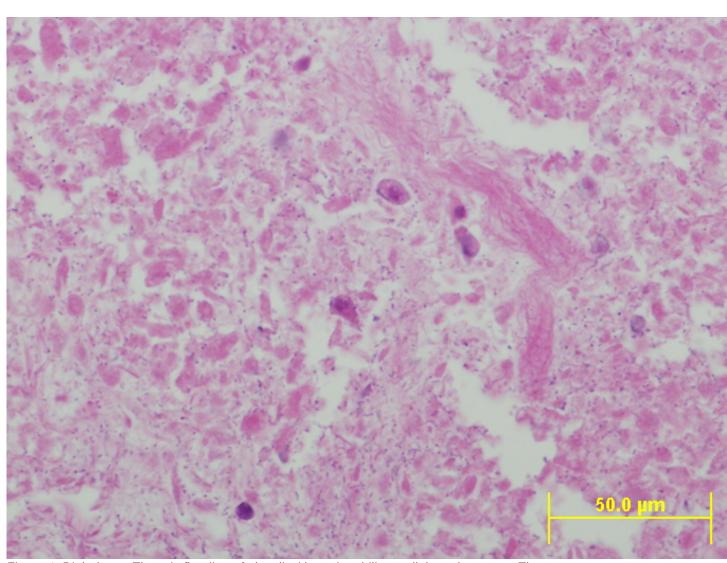


Figure 4. Right lung. There is flooding of alveoli with eosinophilic, acellular substance. There are macrophages (cells with nuclei) present within this eosinophilic substance. Small basophilic stippling represents bacterial populations (post-mortem overgrowth).

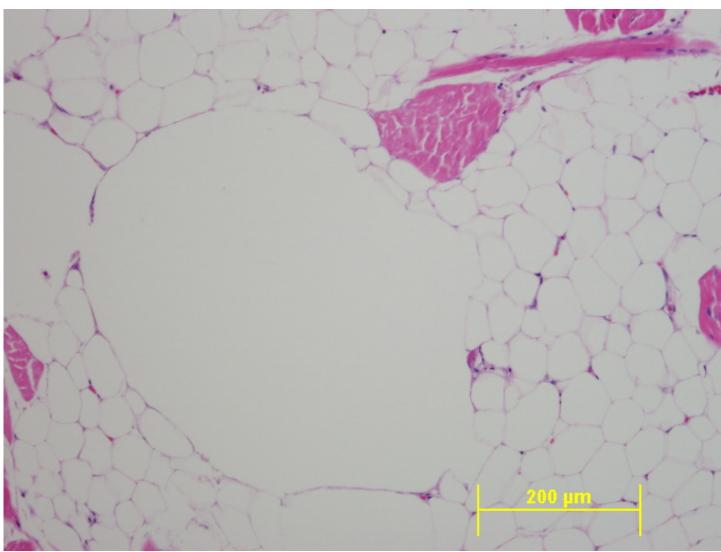


Figure 5. Doral Defect. There are occasional large spaces within the fibroadipose where adipocytes have lost cellular membranes and form a larger structure.

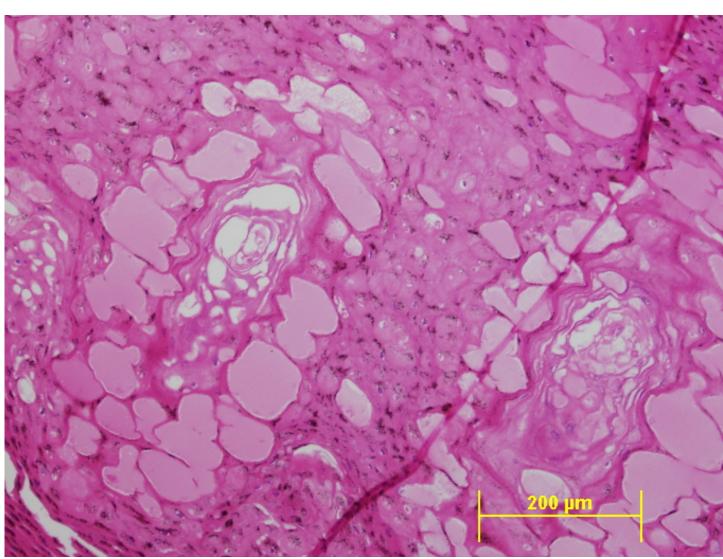


Figure 6. Dorsal Defect. There is accumulation of intracellular eosinophilic substance (serum) within the stratum externum.

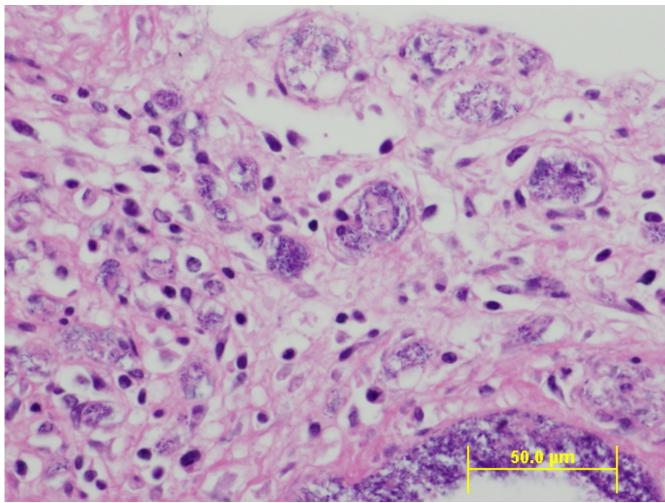


Figure 7. Station I. There are mixed inflammatory cells within the edematous dermis composed of macrophages, lymphocytes, and plasma cells.

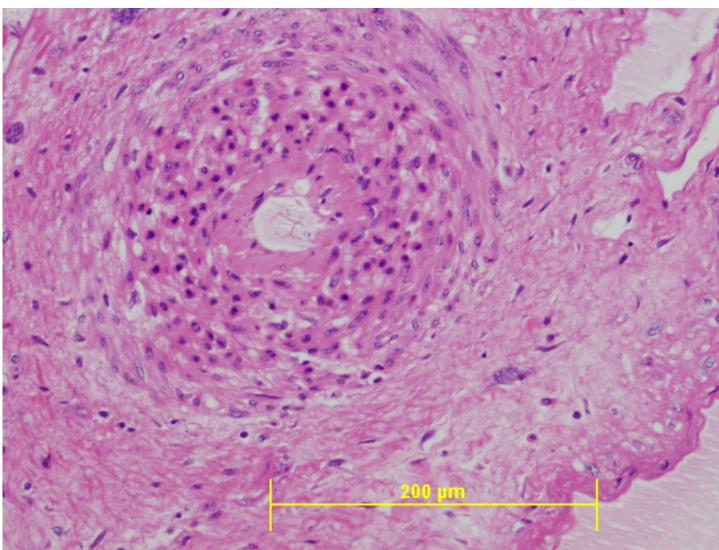


Figure 8. Station I. Within the dermis, there is a well-demarcated granuloma composed of epithelioid macrophages surrounding a core of macrophages and degenerate neutrophils and a central core of eosinophilic substance and few bacteria.

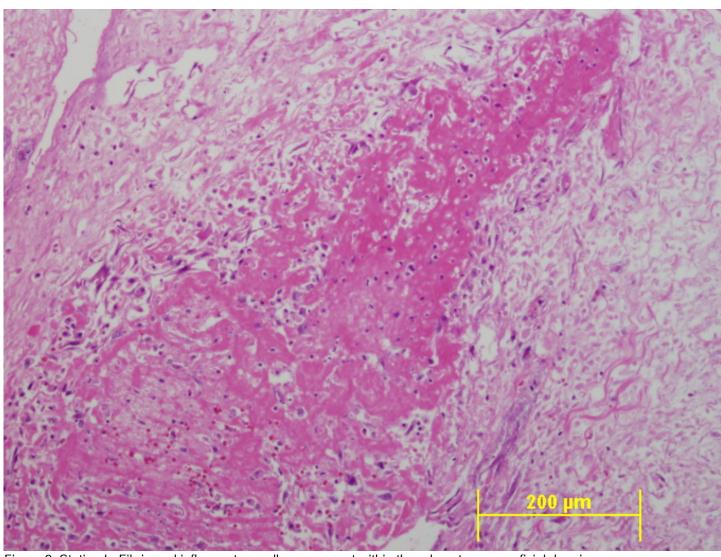


Figure 9. Station I. Fibrin and inflammatory cells are present within the edematous superficial dermis.

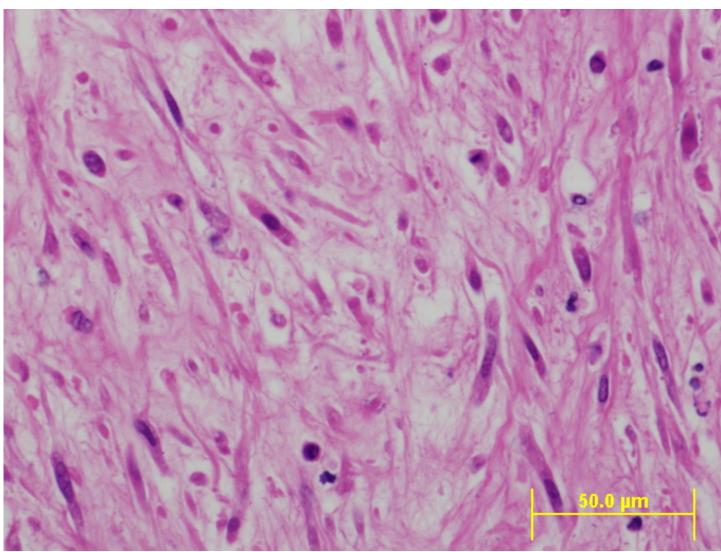


Figure 10. Station I. Fibroblasts are typically loosely arranged. Inflammatory cells are interspersed within this mesenchymal matrix.

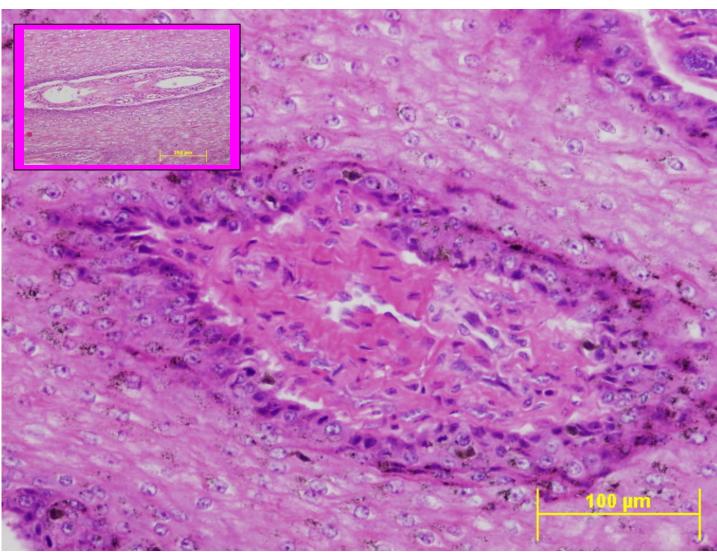


Figure 11. Station H3. Compared to papillary dermal vessels at another site (inset), there is moderate thickening of these papillary dermal vessels and plumpness of lining endothelial cells.

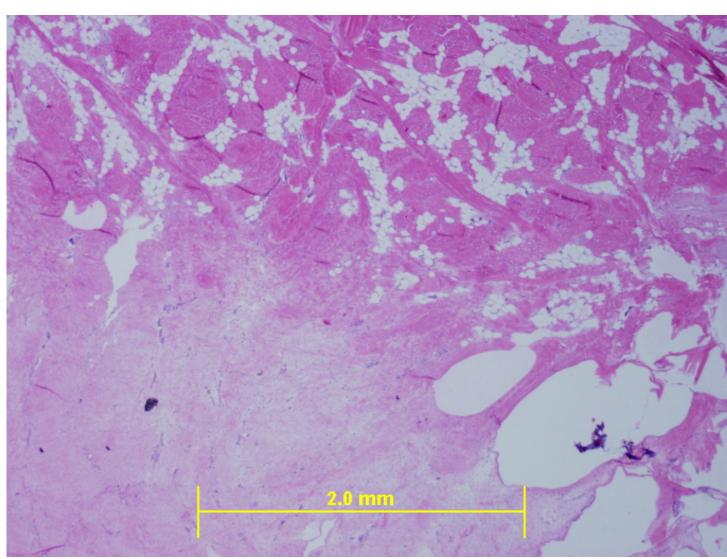


Figure 12. Station H2. Early granulation tissue bed.

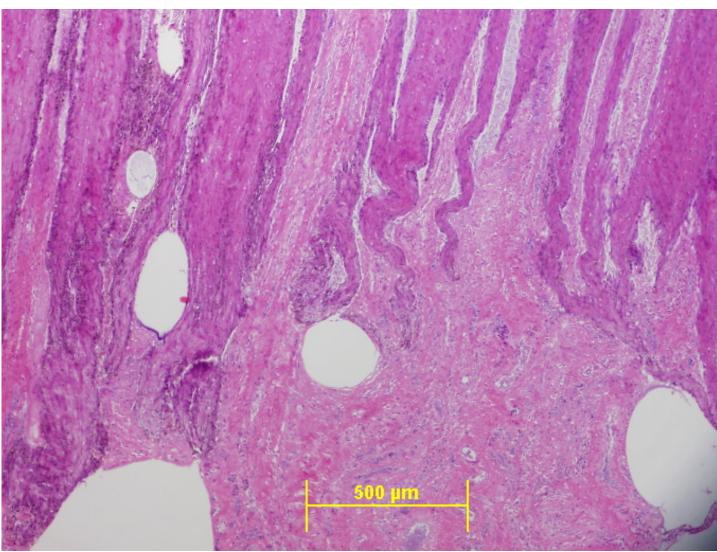


Figure 13. Station H. On the left, there is pigmentation of the epithelium which is decreased to absent on the right (leukoderma).

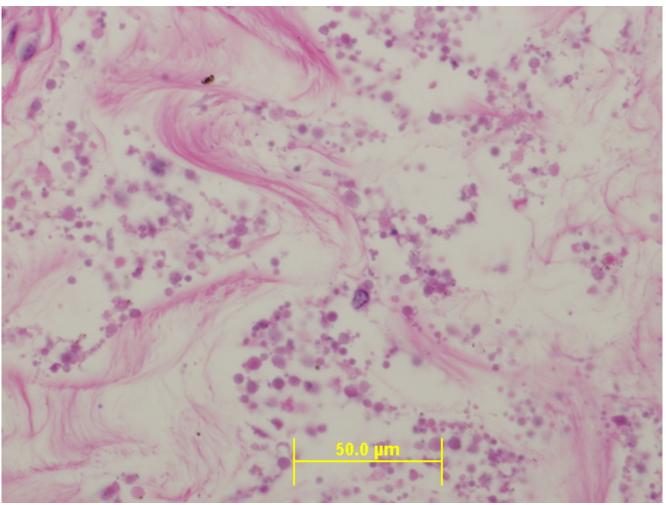


Figure 14. Station H. There is an accumulation of amphophilic, irregular round structures within the dermis. The origin of these structures is not known at this time, but may represent degenerate cells.

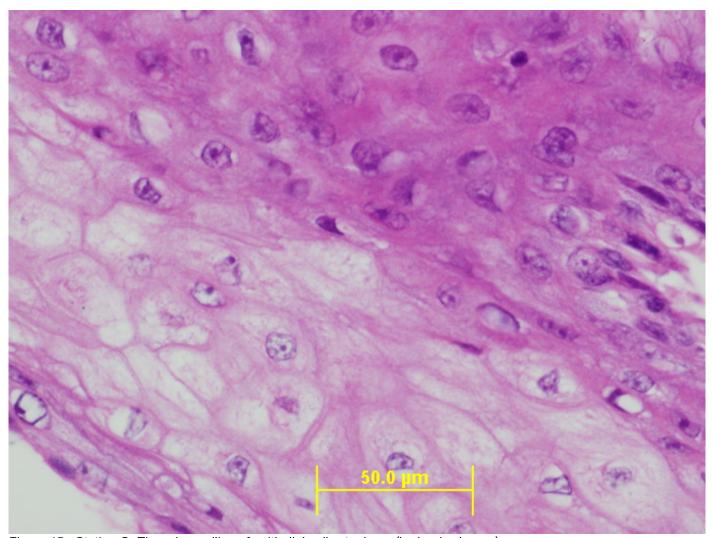


Figure 15. Station Q. There is swelling of epithelial cell cytoplasm (hydropic change).

PENDING TESTS-PATHOLOGIST				
TEST	PURPOSE	SITE	Testing Site	RESULT
PCR-Morbillivirus, Herpesvirus	Viral Presence	Lung, liver, ovary, adrenal gland	OADDL	NEG
Viral Isolation	Viral Presence	Lung, liver, ovary, adrenal gland	NVSL, OADDL	Pending
Brucella	Brucella	Ovary	Mystic	Pending
Special Stains for bacteria (Gram), fungus (GMS), protozoa (Giemsa), and mycobacteria (AFB)-slide 31 only	Infectious agents	Integument	UTCVM	Pending

David S. Rotstein, DVM, MPVM, DACVP Lisa L. Farina, DVM, DACVP REPORTING PATHOLOGISTS

Final Slide Evaluation:6 Feb 2006 Final Summary Date: 6 March 2006

CASE SUMMARY

GROSS OBSERVATIONS (from Final Gross Report)

Integumentary System:

A. Dorsum: Regionally extensive epidermal defect with cranial and caudal irregular and roughened edges and segmental smooth edges without associated subdermal edema or hemorrhage.

B. Peduncle:

- 1. Regionally extensive epidermal and dermal indentation and penetration with overall pattern formation of diamond, vee, and straight lines (net entanglement, vs. shark bite, vs. both) with localized tissue reaction.
- 2. Multiple semi-circular cutaneous defects (shark bite)
- 3. Cutaneous Cyamidiasis, severe.

Internal organs:

Unremarkable to the extent observation was possible given the significant autolysis.

HISTOPATHOLOGICAL OBSERVATIONS (from Final Histological Report)

There was minimal inflammation within the dermis of sections from the dorsal defect and minimal edema. Hemorrhage was not observed. All peduncle lesions were characterized by orderly formation of granulation tissue and all had focal to regionally extensive epithelial lesions (ulceration). Granulation tissue formation is characterized by edema with inflammatory cells and loose fibroblasts transitioning to more organized densely packed fibroblasts interspersed with vascular channels. In one section (I), inflammation was more pronounced and the dermis contained a granuloma. In another section (L), there were superficial intradermal, herniated nests of epithelial cells (suspected traumatic implantation). Epithelial hyperplasia was present in all peduncle sections examined.

INTERPRETATION

See case summary in Histopathology Report pages 4 and 5

CAUSE OF DEATH

Open, but given the apparent pre-mortem shark and net entanglement damage to the peduncle, in the absence of any other significant information, the most parsimonious hypothesis is that these injuries were sufficiently serious to initiate the demise of the case.

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